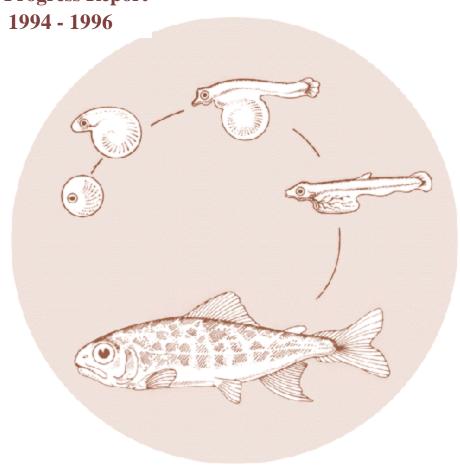
# Study to Determine the Biological Feasibility of a New Fish Tagging System

# **Progress Report**





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# A STUDY TO DETERMINE THE BIOLOGICAL FEASIBILITY OF A NEW FISH TAGGING SYSTEM, PART I:

# **Evaluation of Potential Passive Acoustic Tag Systems**

## PROGRESS REPORT 1994-1996

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#### **EXECUTIVE SUMMARY**

We investigated the technical feasibility of developing two types of passive acoustic tags, each with a read range of up to 10 m. The first tag evaluated was an acoustic passive integrated transponder (A-PIT) tag. Our initial evaluation addressed the attenuation of acoustic energy as it propagates through the body of a fish in order to determine the range of viewing aspects from which tags could be effectively read. A test transducer was placed in the coelomic cavity of fish, and results showed a wide range in attenuation values for the frequencies of interest (50 and 500-kHz) in relation to aspect or viewing angle.

The data suggested that a fish's body and air bladder would significantly attenuate acoustic signals at most viewing angles except ventrally and to a limited extent laterally. In addition, due to the narrow dimensions of the beam at short range, fish rapidly passing near the energize/receive transducer may not remain within its interrogation field long enough for the tag to be energized or for the return signal to be received completely.

Two separate sound fields were proposed for use with the A-PIT tag system: one at 50 kHz (a continuous energizing field) and the other at 500 kHz (tag response frequency). The strengths of the 50- and 500-kHz sound fields were estimated at 207-213 dB $\mu$ Pa@1 m and 120-126 dB $\mu$ Pa@1 m, respectively. Thus, during operation of an A-PIT system, fish and other animals could be exposed to strong sound fields. A literature review showed that the energy field required to energize the A-PIT tag could, under some conditions, cause behavior modification and/or damage some animals.

We concluded that technically, the A-PIT tag could be developed. This was confirmed by an independent non-government contractor who reviewed the potential system. However, based upon the signal attenuation data showing limited operational viewing aspects, and the literature review showing a potential risk to fish and other animals from the continuously transmitted tag-energizing field under some potential operating conditions, we recommend that the tag not be developed. These factors, in addition to potentially high system developmental cost, outweigh any potential advantages of the system over currently used tagging systems.

Similarly, upon investigation of the technical feasibility of a resonating sphere tag, it became apparent that the tag could be developed (confirmed by an outside non-government contractor) but that its application would be limited, and there would be a potential risk to animals confined in close proximity to the tag energizing system.

Factors identified as potentially limiting system performance included use with small fish only, diminishing tag-detection ability as a fish grows, limited "tag codes" (resonating frequencies) because of tag size limitations in relation to fish size, ambient noise reducing tag-detection ability, limited viewing aspects because of a fish's physical characteristics, and the acoustic spectral characteristics of fish.

Calculations showed that the strength of the acoustic energizing field for a resonant sphere tag system could, as with the A-PIT tag, potentially cause behavior modification or damage to fish or other animals. Based on this information, we recommend that development of a resonating acoustic sphere tag for use in the Columbia River Basin not take place at this time.

A review of literature covering the effect of sound on animals strongly suggested that the interrogation or insonification sound-field strength of either of the proposed tag detection systems could, under certain conditions, cause harm and/or behavior modification to fish and aquatic mammals. However, the ability of some animals to detect and avoid a potentially damaging sound field prior to damage taking place reduces this concern. To reduce the risk of harm, the systems would thus need to be operated in situations that do not confine animals (i.e., use in open water and not in fish ladders).

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# INTRODUCTION: EVALUATION OF POTENTIAL PASSIVE ACOUSTIC TAG SYSTEMS

Fisheries agencies use a variety of marking systems to identify groups or individual fish. One such system is the radio frequency passive-integrated-transponder (RF-PIT) tag, which obtains its operating power from an electromagnetic field (EMF) and subsequently "transmits" its unique identification code to a receiver (Prentice et al. 1990). The major shortcoming of this passive tag is its limited operating range.

In 1992, NMFS proposed to Bonneville Power Administration (BPA) that the technical feasibility of developing a new generation of small passive tags and tag detection systems, suitable for use with juvenile salmonids, be investigated. As proposed, the tags would operate using acoustic rather than electromagnetic energy, thus increasing their detection and read range over existing tags. Theoretically, these systems would be able to detect acoustic tags to a range of up to 10 m or more.

The investigative work took place during the period of 1994 through 1996 as a work element within Project 83-319, "A Study To Determine The Biological Feasibility Of A New Fish Tagging System, Part III: Development and Evaluation of PIT-tag Technology." Two types of passive sonic tags and the potential effect of acoustic energy (acoustical field intensities proposed to energize tags and transponder tag data) on biota were investigated and are discussed in three separate papers within this report.

#### TECHNICAL FEASIBILITY OF AN ACOUSTIC PIT-TAG

#### Introduction

The first tag the NMFS proposed to investigate was an acoustic passive integrated transponder (A-PIT) tag. As proposed, the tag would have the following features:

- Passive (i.e., having no power source of its own)
- An operating range of up to 10 m
- Energized with one acoustic frequency and transmit data on another
- Individually coded
- Suitable for implanting into the coelomic cavity of juvenile salmonids

A multi-phased plan to investigate the technical feasibility of such a tag was formulated. Since the proposed tag would lie in the coelomic cavity of a fish, its design necessitated that it be energized and respond through the fish body. Thus, the first steps in the investigation were to determine acoustic energy attenuation through the bodies of fish to estimate acoustic power levels needed to operate and detect the tag, characterize the limitations of the tag performance, and to develop a prognosis of the applicability of the tag for field research.

#### **Methods and Materials**

In this study, attenuation of acoustical energy through fish bodies was measured for body aspect angles throughout pitch, roll, and yaw planes (Table 1, Fig. 1). Attenuation for each aspect (viewing) angle was unique with respect to the tissues and structures encountered along that path of sound propagation. Factors affecting attenuation included how fully the swim bladder was inflated, the thickness and composition of bone and musculature, and frequency-dependent effects. Attenuation was measured using a small test transducer (underwater speaker/microphone) that was inserted into the coelomic cavity of fish at approximately the position where an injected A-PIT tag would lie.

Calibrated laboratory transducers were used to transmit to and receive transmissions from the test transducer. Measurements were made using two frequencies, 50 kHz and 500 kHz. Fifty kHz was chosen to represent the tag energizing frequency. Acoustical theory predicts that attenuation through a fish body will be less for 50 kHz than for a higher frequency, which should facilitate energy transfer to a tag.

Table 1. Definitions of the three planes of rotation (with angular references, Fig. 3a-f) used in the study of acoustical attenuation by a fish's body.

Plane of Rotation	Description
Pitch	The plane that divides a fish or the transducer into left and right halves (sagittal plane) (tail-on = 0°, ventral aspect = 90°, head-on = 180°, dorsal aspect = 270°; Figs. 3b, 3e)
Roll	The plane that passes through and normal to the long axis and divides a fish or the transducer into fore and aft halves (transverse or orthogonal plane) (dorsal aspect = $0^{\circ}$ , ventral aspect = $180^{\circ}$ , side aspect = $90^{\circ}$ and $270^{\circ}$ ; Figs. 3a, 3d). The roll plane intersected the body of a fish near the insertion of the dorsal fin.
Yaw	The plane passing parallel to the lateral lines that divides a fish or transducer into upper and lower halves (or median longitudinal plane) (tail-on = $0^{\circ}$ , head-on = $180^{\circ}$ , side aspect = $90^{\circ}$ and $270^{\circ}$ ; Figs. 3c, 3f). This configuration was indistinguishable from the pitch plane for the (cylindrical) transducer when viewed alone since its directivity was nearly symmetrical about the roll axis.

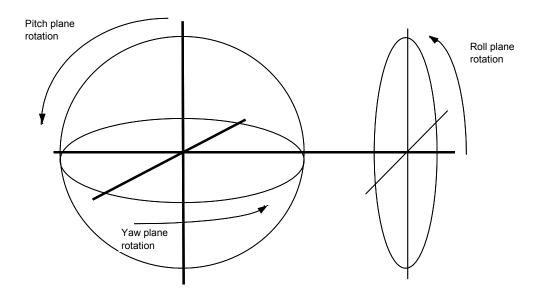


Figure 1. Pitch, roll and yaw planes of rotation used for measurement of baseline transducer directivity and acoustical attenuation effects of fish body structure. Fish were mounted on the horizontal bar, dorsal side up, and facing to the left.

Further, the diameter of a transducer capable of producing the necessary interrogation field intensity would be of a manageable size; less than 10 cm (Kinsler et al. 1982). Five hundred kHz was chosen to represent the tag response frequency because of the necessarily small transducer that would be incorporated into an A-PIT tag. The physical size of a transducer element is inversely related to its resonant frequency (Kinsler et al. 1982). In addition, a high frequency allows a greater rate of tag-code data transfer.

All measurements for this study were made on the University of Washington's acoustics barge (*R. V. Henderson*) while it was moored in 4.3 m of fresh water. The *Henderson* is a self-contained, floating laboratory for underwater acoustics research. It houses a full range of electronic test and measurement equipment, including calibrated transducers (hydrophones). Water temperature throughout the investigation was 7.5°C.

The test transducer used in this study was not calibrated, so it was not practical to attempt to directly measure acoustical attenuation through a fish body. Thus, comparative measurements were made to determine the reduction in acoustical pressure attributable to attenuation. To make these calculations, the directivity (radiation or receiving sensitivity pattern) of the test transducer was measured alone, and measured again when inserted into a fish. A particular set of aspect angles with respect to the physical axes of the transducer were maintained throughout both sets of measurements.

This comparative procedure accomplished two objectives. First, the differences between the two measurement sets constituted a direct measurement of attenuation. Secondly, most of the basic directivity effects of the test transducer were eliminated because only changes in acoustical pressure at particular aspect angles were used to calculate attenuation. Thus, test transducer directivity effects were removed from fish body attenuation measurements. A test transducer having an omnidirectional directivity pattern would have been desirable for these measurements. However, the design of such a transducer is very difficult, and its construction would have been prohibitively expensive.

The test transducer was constructed by the Applied Physics Laboratory (APL) at the University of Washington. It consisted of a single, hollow ceramic cylinder, 5.1-mm long by 2.5-mm in diameter. The finished unit measured 5.6-mm long by 4.0-mm in diameter (after encapsulation) and was attached to a 3.0-m length of RG-147 coaxial cable. Voltages representing acoustical field strengths striking the test transducer were measured at the end of the coaxial cable. Transmissions from the test transducer were measured by calibrated transducers located at a fixed horizontal range of 1.57 m.

All directivity measurements were made either with the test transducer alone or with the test transducer inserted into the coelomic cavity of a fish mounted within an adjustable suspension frame (Fig. 2). The frame was mounted on a rotatable shaft that was lowered to position the test transducer at a 2.1-m depth. Test subjects were positioned within the frame such that when rotated horizontally, a line between the test transducer and the calibrated laboratory transducer described a desired aspect plane (i.e, pitch, roll, or yaw).

The frame consisted of two pairs of metal supports, with each pair fastened together to form symmetrical crosses suspended 2-m apart, one above the other. The supports were suspended using 0.75-mm-diameter, plastic-coated, stainless steel wire. The upper members of the frame were made of 1.2-m lengths of 1.25-cm inside-diameter steel pipe, while the lower members were made of similar lengths of 7.6-cm-wide by 6.0-mm-thick steel flat-bar. The wider dimension of the flat-bar was placed parallel to the water surface. Steel flat-bar was used for the lower support because its additional weight and small cross-sectional area aided in stabilizing the frame when it was submerged and rotated.

Pitch- and roll-plane directivity data sets were collected for the test transducer alone for use as baseline measurements prior to making measurements with the transducer inserted into a fish. Directivity was nearly symmetrical throughout the roll plane of the test transducer, so pitch- and yaw-plane directivity were considered as equivalent. When the test transducer was inserted into the coelomic cavity of fish, its long axis was aligned as closely as possible with the long axis of the fish. Directivity data were then collected throughout pitch, roll, and yaw planes with respect to the axes of the fish (Fig. 1).

Freshly sacrificed fish of two general size groups were used as test specimens. The smaller individuals were sockeye salmon (*Oncorhynchus nerka*) that ranged in fork length from 24.8 to 26.7 cm. The larger fish were Atlantic salmon (*Salmo salar*) that ranged in fork length from 54.6 to 61.0 cm. A fish and/or the transducer was suspended within the test frame using monofilament fishing line (1.8 or 5.5 kg breaking strength depending upon fish size) and fishing hooks (#8-10).

The test transducer was inserted into specimen fish through a 3-mm-long incision in the abdominal musculature located posterior to the pectoral fins and 3 to 5 mm from the mid-ventral line. The coaxial cable leading from the transducer was routed through the opercular slit and out the mouth. Air or water intrusion was reduced by application of petroleum jelly around the coaxial cable entry incision. Fore and aft adjustments of the transducer within the coelomic cavity were made using a reference mark on the coaxial

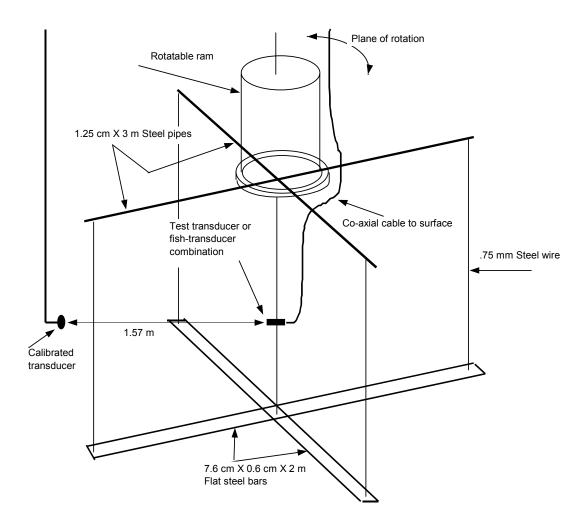


Figure 2. Diagram of the test frame used for measurement of baseline test transducer directivity and acoustical attenuation effects of fish body structure. Fish were mounted within the frame so that when rotated in the horizontal plane, their pitch, yaw or roll planes were presented to the calibrated transducer.

cable. However, dorso-ventral and side-to-side misalignments could not be observed directly and were estimated by the angle at which the cable emerged from the incision.

The test transducer was placed adjacent to the abdominal musculature at the ventral surface of the coelomic cavity, about 2 cm forward from the pelvic girdle. This is the normal location where an A-PIT tag would be placed within a fish. Attenuating effects of air in swim bladders were measured with swim bladders fully inflated, partially inflated, and deflated.

Air volumes were adjusted using a 60-cc syringe attached to a 20.5-mm long, 22-gauge hypodermic needle. The needle and syringe were attached by a 3.0-m length of 1.8-mm (outside diameter) plastic tubing. The needle was inserted into the air bladder and the tubing was routed forward from the needle through the opercular slit and out the mouth to the surface. The hypodermic needle was left in position during all measurements, and the tubing was positioned for least interference to acoustic measurements.

All acoustic measurements consisted of two types. The first type was of the acoustical pressure received by the test transducer from a fixed source at uniformly spaced aspect angles, either alone or within a fish, as it was rotated through 360°. The second type of measurement was of the acoustical pressure received by a calibrated measurement hydrophone, which was radiated by the test transducer at similar aspect angles.

Receive measurements were made with a constant 50-kHz acoustic field level directed toward the test transducer using a calibrated transducer positioned at a fixed location. Similarly, a calibrated transducer at the same location was used to measure the strength of a 500-kHz acoustical field radiated from the test transducer, which was energized by a constant input level.

Measurement values were recorded in decibel units, relative to one micro Pascal ( $dB\mu Pa$ ). Directivity data sets, as used in this report, consist of groups of acoustical pressure measurements received by or transmitted from the test transducer at incremental angles throughout 360 degrees of rotation within a particular plane. Total attenuation by a fish was calculated as the sum of 50 kHz (simulated tag interrogation field) and 500 kHz (simulated tag transponding field). Attenuation effects differ for the two frequencies when they propagate through different types of tissue (see Kinsler et al. 1982 for a detailed review of frequency-dependent attenuation of sound propagation).

#### **Results and Discussion**

With the test transducer implanted within the coelomic cavity of a fish, most measurements of sound attenuation were characterized by wide ranges in values with respect to aspect or viewing angle. The largest source of variability in measured attenuation was related to the physical properties of a fish. These properties are of primary interest since they suggest aspect angles where an implanted A-PIT tag could possibly be detected.

Most repeated directivity data sets for a particular test transducer/fish configuration were similar when no physical adjustment of the fish or transducer was made between measurements. However, physical adjustments (i.e., repositioning of the test transducer or fish on the test frame; lifting the frame out of the water and repositioning it at depth) often resulted in changes in measured directivity. The frame was not sufficiently rigid to maintain or allow precise realignment of the transducer or of a fish. In addition, large differences in directivity values were seen between fish. This resulted primarily from size differences between specimens and individual acoustical characteristics of fish.

Two factors unrelated to the characteristics of fish were identified as contributing to the variability between attenuation measurements. They were misalignment of test transducer/fish combinations on the test frame and uncertainty in swim-bladder inflation. Due to its physical shape, the directivity (radiation and receiving sensitivity patterns) of the test transducer varied considerably with aspect. Likewise, the acoustical attenuating characteristics through a fish body varied for different aspects, depending on the organs and structures through which sound propagated. Thus, moderate misalignments of the test transducer and the fish could result in large apparent changes of attenuation, particularly where large changes in test-transducer directivity occurred over small changes in aspect angle, since directivity effects are additive.

Acoustical theory predicts high attenuation of sound propagation through an air bubble equivalent to the size of a fish swim bladder (Kinsler et al. 1982). However, no clear trend was seen in the degree of swim-bladder inflation and acoustical attenuation during this study. Overall, attenuation was less when the swim bladder was deflated. However, attenuation effects of the swim bladder were not significantly greater than those of the heavy dorsal musculature and skeletal structure of a fish.

Complete deflation of the swim bladder was difficult to achieve. Dissection revealed that the swim bladder did not collapse evenly or fully using the syringe. In all observed cases, some air remained trapped in parts of the swim bladder. Surgical deflation of the swim bladder required that the transducer be removed while the coelomic

cavity was opened and the swim bladder was punctured and completely evacuated of air. In addition, lifting the test frame out of the water, and later lowering it back to depth, caused considerable inadvertent misalignment of the transducer and fish, contributing to uncertainty in comparing attenuation measurements.

Although the fore/aft location of the test transducer was known, small changes in its location relative to the pelvic girdle had not been predicted to significantly affect acoustical attenuation. However, for the larger fish, considerably higher attenuation was noted when the internal transducer was positioned about 2 cm forward of the pelvic girdle than when placed directly dorsal of the girdle. The effect probably was related to differences in the size and shape of the swim bladder at the two locations and perhaps to differences in its proximity to the test transducer.

Dorsal aspect attenuation of acoustical pressure amounted to 25-30 dB reduction (94-97%) for 50 kHz and 15-20 dB reduction (82-90%) for 500 kHz, when the test transducer was forward of the pelvic girdle. However, when the transducer was directly dorsal of the pelvic girdle, attenuation was only a 5- to 10-dB reduction (44-68%) for either frequency. Similar measurements were not made using the smaller size fish. It is likely, based on our experience with RF-PIT tags, that under normal conditions some A-PIT tags could migrate between either location. Those positioned just dorsal of the pelvic girdle would be subjected to lower attenuation effects and would have a higher probability of detection.

The sum of acoustical attenuation of 50-kHz energization and 500-kHz response fields through a fish body will determine at which aspect angles an internal A-PIT tag would be detectable. Generalized degrees of attenuation with respect to aspect angle were compiled by comparison of attenuation plots for all fish-size and swim-bladder inflation conditions tested. Predictions of the ranges of aspect angles over which A-PIT tags would be reliably detectable were made using a composite of this data.

These predictions are based on the assumption that for a 10-m maximum range, tag detection would be unlikely if total (2-way; energization + response) fish-body attenuation was more than 24 dB (94% reduction). This somewhat arbitrary threshold was chosen to reflect the source level and sensitivity of a typical echo-sounding system and a practically attainable tag-response source level. The aspect angles where attenuation was greatest for large and small fish were similar. High attenuation values were common in pitch and roll planes to at least 45 degrees fore/aft or left/right from the dorsal aspect. In addition, high attenuation was consistent within 10-20 degrees of the tail-on aspect.

Attenuation at the near head-on aspect was less than at the tail-on aspect, but was still sufficiently severe that the likelihood of reliable A-PIT tag detection would be marginal. Qualitative predictions of pitch, roll, and yaw plane aspect angles where A-PIT tagged fish would most likely be detectable are given in Table 2. Attenuation plots typical of those used to predict reliable tag detection are shown in Figures 3a-f. Attenuation, when presented as a polar plot, shows variations with aspect angle throughout a plane of rotation. Attenuation levels of 0, -6, -10, and -20 dB are equivalent to sound pressure reductions of 0, 50, 69, and 90%, respectively. Note that the plots in Figures 3a-f are only typical of directivity measurements and may not correspond precisely with the ranges of predicted tag-detection ability in Table 2.

Even though there was considerable uncertainty in the measurements made during this study, it was shown that high acoustical attenuation was typical over much of the dorsal aspect of fish for all sizes tested. Attenuation levels were shown to be severe enough to significantly limit the usefulness of an A-PIT tag detection system using a downward directed interrogation beam pattern. Therefore, installations of interrogation and receiving transducers on the hull of a vessel would probably not yield reliable results.

Conversely, low levels of attenuation were found over most near-ventral aspect angles, showing that an upwardly directed acoustical beam could probably detect tags with acceptable reliability. Transducers mounted on the floor of a fish ladder or on the bed of a stream would probably yield satisfactory results. However, if the fish routinely approached close to the transducer, tag detection could be difficult due to the narrow width (small sampling volume) of the beam at very short range.

Acoustical attenuation through a fish body is not the only factor that would limit the detectability of A-PIT tags. Ambient noise and bubbles created by wind, turbulence, waterfalls, etc. would interfere with tag detection in at least two ways: acoustical noise would obscure recognition of some tag responses, and bubbles would attenuate sound fields traveling to and from a tag. It is not known how sensitive A-PIT tag decoding would be to environmental noise interference. Therefore, custom installation would be required at each interrogation site to overcome these potential problems.

An independent review of the data sets collected during this investigation was made by GRD Associates (1994, Appendix A). The author concluded that development of a system that could reliably detect implanted A-PIT tags to a range of 10 m could be accomplished, but concurred that there are aspect angles in which a tag may not be detectable.

Table 2. Predicted ranges of aspect angles where one-way acoustical attenuation through the body of a salmonid would allow A-PIT tag energization (50 kHz) or tag response detection (500 kHz). Note that total (two-way) attenuation equals the sum of energization plus response frequency attenuation. A-PIT tags would be detectable only within overlapping angle ranges for the two frequencies, for each aspect plane listed below. Angles are referenced to those in the attenuation plots shown in Figure 3a-f.

	Frequency		
	50 kHz	500 kHz	
Viewing aspect	(range)	(range)	
D 1			
Dorsal aspect	a	a	
Pitch plane	a	a	
Roll plane			
Ventral aspect			
Pitch plane	30°-190°	55°-125°	
Roll plane	85°-275°	80°-280°°	
_		40°-320° <sup>d</sup>	
side aspect			
Yaw plane	30°-150°	40°-140°	
Roll plane	10°-180°	10°-180°	
lead-on aspect			
Pitch plane	140°-200°	b b	
Yaw plane	150°-210°		
Tail-on aspect			
Pitch plane	10°-200°	b b	
Yaw plane	20°-340°		

a Undetectable for > 45 degrees.

b Could not be reliably determined due to nulls in the test transducer directivity pattern at this particular aspect.

c Inflated swim bladder.

d Deflated swim bladder.

GRD Associates presented several suggestions for improving tag-detection ability. These included design of a tag with increased transmitting power, design of an active-when-interrogated tag using an internal battery to supplement transmitting power, and increased tag-energization field strength. GRD Associates also suggested that tag detection would be more reliable if interrogator receiving sensitivity could be increased and the maximum required operating range (10 m) were reduced. Implementation of some of these suggestions would change various characteristics of the A-PIT tag as currently envisioned. The tag would have to be larger to contain an internal battery, developmental costs would be probably increase, and the interrogation system would be more complex.

Based on measurements of acoustic field attenuation by fish bodies, and the independent review of the results of our measurements, we recommend that development of an A-PIT tag detection system not be undertaken at this time. The aspect angles for viewing the tag are too restrictive for broad research application in the Columbia River Basin. In addition, environmental noise would further complicate operation of the tag system in most areas of interest.

# Roll Plane Directivity; 50 kHz

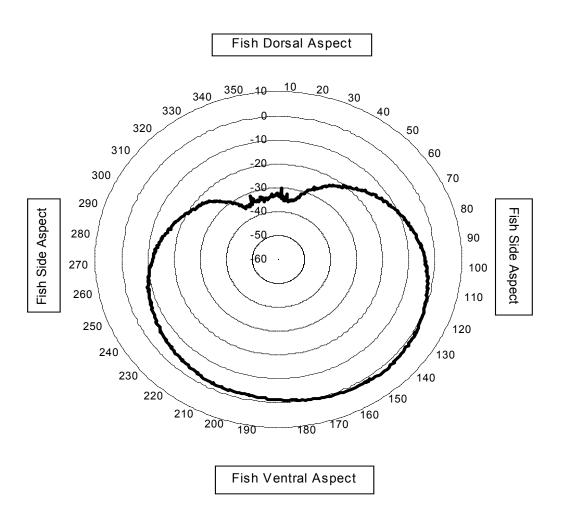


Figure 3a. Typical directivity plot of roll plane acoustical attenuation due to a fish body for 50 kHz; measured with the test transducer inserted into the coelomic cavity of a fish, at a location where an A-pit tag would be expected to lie.

# Pitch Plane Directivity; 50 kHz

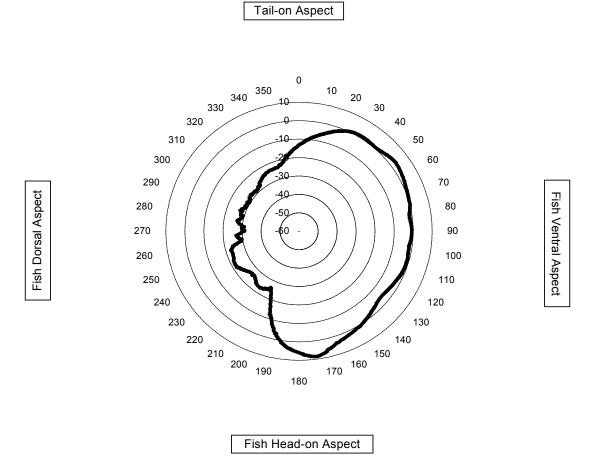


Figure 3b. Typical directivity plot of pitch plane acoustical attenuation due to a fish body for 50 kHz; measured with the test transducer inserted into the coelomic cavity of a fish, at a location where an A-pit tag would be expected to lie.

# Yaw Plane Directivity; 50 kHz

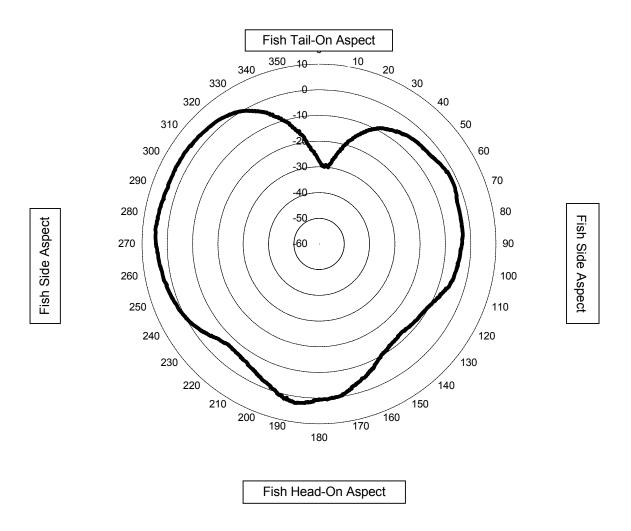


Figure 3c. Typical directivity plot of yaw plane acoustical attenuation due to a fish body for 50 kHz; measured with the test transducer inserted into the coelomic cavity of a fish, at a location where an A-pit tag would be expected to lie.

# Roll Plane Directivity; 500 kHz

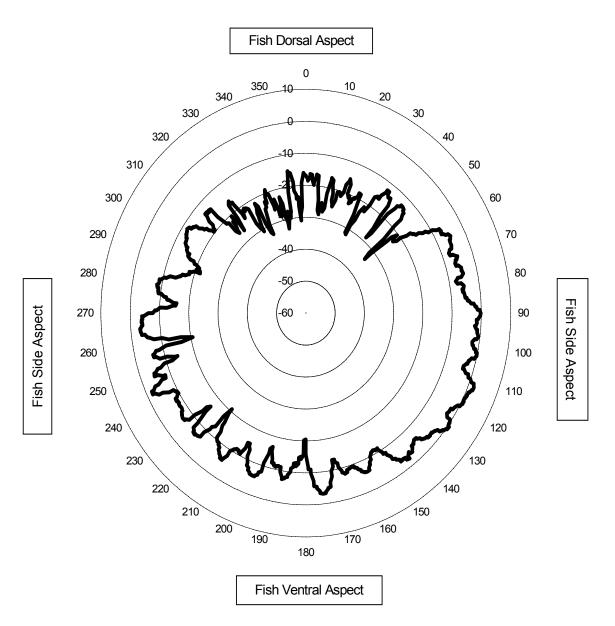
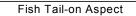


Figure 3d. Typical directivity of roll plane acoustical attenuation due to a fish body for 500 kHz; measured with the test transducer inserted into the coelomic cavity of a fish, at a location where an A-pit tag would be expected to lie.

# Pitch Plane Directivity; 500 kHz



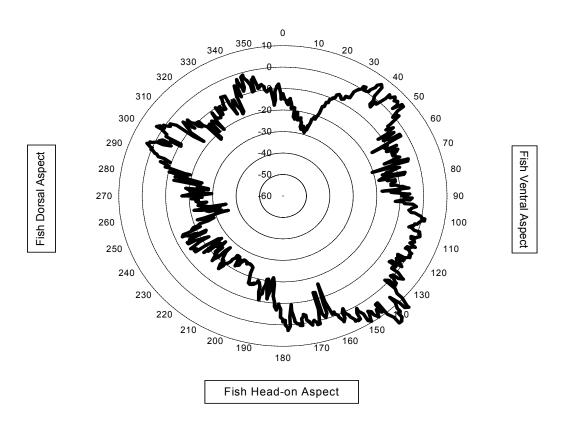


Figure 3e. Typical directivity plot of roll plane acoustical attenuation due to a fish body for 500 kHz; measured with the test transducer inserted into the coelomic cavity of a fish, at a location where an A-pit tag would be expected to lie.

# Yaw Plane Directivity; 500 kHz

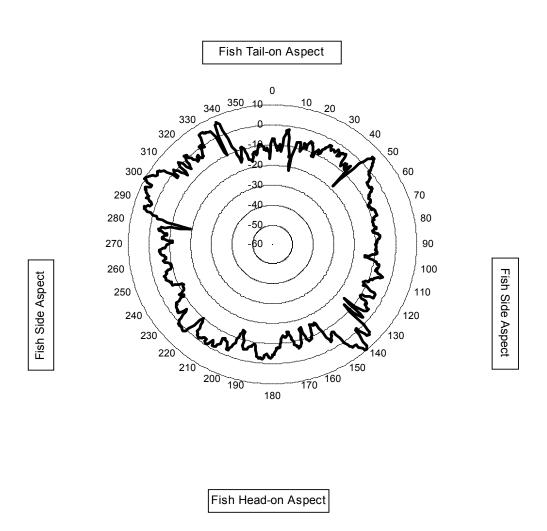


Figure 3f. Typical directivity plot of yaw plane acoustical attenuation due to a fish body for 500 kHz; measured with the test transducer inserted into the coelomic cavity of a fish, at a location where an A-pit tag would be expected to lie.

#### **Conclusions and Recommendations**

- 1) Attenuation of 50- and 500-kHz frequencies by the bodies of differing sizes of salmonids was investigated.
- 2) Calculations were based on the assumption that the tag would operate at a range of up to 10 m from the interrogating/receiving transducer.
- 3) All measurements of attenuation were made by comparison of acoustical field strengths received or generated by a test transducer operated both outside and surgically implanted in the coelomic cavity of a fish, at similar aspect angles. The test transducer alone or the fish with the implanted test transducer were positioned on an adjustable support apparatus during all measurements.
- Wide ranges in attenuation values, in relation to aspect or viewing angle, were observed. Measurement variability was attributed to the following factors, in order of importance: viewing or aspect angle, fish size, location of the transducer within a fish, air-bladder inflation variability, and physical test apparatus.
- 5) High attenuation values were common to at least 45 degrees from the dorsal aspect of the fish in all directions.
- 6) High signal attenuation within 10-20 degrees of the tail-on aspect was observed.
- 7) Attenuation at the near head-on aspect was less than at tail-on aspect but was still marginal for reliable detection.
- 8) Low attenuation was observed for all fish for the near-ventral aspect angles.
- 9) Short-range ventral viewing may be difficult because of acoustic beam narrowing very near the transducer.
- 10) Results suggest that an upward- and lateral-viewing A-PIT tag system could detect tags with acceptable efficiency.
- Ambient noise will reduce tag detection efficiency because of interference with its weak return signal.
- 12) An independent review of NMFS data by GRD Associates concluded that development of an A-PIT tag was technically feasible, but when injected into a fish would be limited in the range of aspect angles at which it could reliably be

detected. GRD Associates offered several suggestions for improving the tag, including adding a battery to improve tag return-signal strength. However, this approach changes the tag concept from a passive to an active tag, and as such the tag would have a limited operating life.

We concluded, because of the limited viewing aspect angles where tags could be reliably detected and the possible limitations imposed by environmental noise, that the tag not be developed.

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## TECHNICAL FEASIBILITY OF AN ACOUSTIC RESONANT TAG

#### Introduction

A marking tool that will allow remote, non-invasive detection and recognition of tagged fish groups is needed for a variety of fisheries research and management programs. Such a tag must be reliable, stable, and must not impose a significant biological burden. A hollow glass sphere that acoustically resonates could potentially be such a tag. Like passive integrated transponder (PIT) tags, the spheres could be implanted into the coelomic cavity of fish. As envisioned, the resonant tags would be detected by spectral analysis of acoustic echo returns from exposing fish to a swept frequency (limited portion of a broad frequency band) pulse (an acoustical chirp), which would include the resonant frequency of the implanted tag.

The frequency range of the chirp could be selected from a large spectrum, perhaps ranging from 10 to 500 kHz, and would be selected depending upon the characteristics of a particular fish/resonant sphere combination of interest. The reflective contribution or "fingerprint" of the resonant tag would give tagged fish a different spectral pattern than untagged fish. Thus, tagged fish and untagged fish of similar size could be distinguished. This approach would permit identifying, enumerating, and determining the distributions of tagged fish.

In concept, a resonant tag is simple and appealing, but it may be difficult to implement because the tag's "fingerprint" could be masked by the spectral characteristics of the fish or by ambient environmental noise. In addition, an effective resonant sphere for implantation into fish and a resonant-tag detection system do not currently exist and would need to be developed. Furthermore, the acoustical field strength needed for acquisition of sphere echo returns from a 10-m range was estimated at 207-213 dB $\mu$ Pa@ 1-m from the face of the transducer. This level of acoustic energy could have a detrimental effect on biota.

Each of these factors needs to be investigated to determine if the effort is justified. In addition, calculations estimating the acoustical reflectivity or target strength (TS) of fish, TS of resonant spheres, and enhancement of fish TS by an implanted resonant sphere are essential in determining the feasibility of the proposed tag. This paper addresses the enhancement of fish TS.

#### **Methods and Materials**

The target strength of an object (i.e., a fish) is calculated as 10 times the logarithm of the ratio of incident to reflected acoustical intensity, at a range of 1 m. By convention, a perfectly reflecting sphere having a radius of 1 m has been adopted as a 0-dB target-strength reference. Since the surface, or back-scattering area of this sphere is  $4\pi$  m<sup>2</sup>, the effective back-scattering area of another reflector can be calculated from its target strength, which is also the ratio of its scattering area to that of the standard sphere. The TS of a fish is directly proportional to its length and indirectly proportional to the frequency of an applied acoustical pressure field (Table 1).

Target strength (in dB), based on scattering area, is defined as follows (Appendix B):

$$TS_{(\sigma)} = 10 \log(\sigma/4\pi) \tag{1}$$

where  $\sigma$  = effective scattering area of a fish (m<sup>2</sup>) and  $4\pi$  = surface area of a 1 m radius sphere.

The target strength of a scatterer such as a fish is related not only to its length, but also to the frequency of the acoustic field used to measure reflectivity. Fish TS may be estimated as below (Appendix B):

$$TS_{(fish)} = 10 \times log(L^{1.87} \times \lambda^{0.13}) - 27.7$$
 (2)

where L = fish length (m) and  $\lambda = acoustic wavelength (m).$ 

The TS of different sized, very thin-walled spheres (thickness ratio = inner radius/outer radius = 0.99), near their resonance frequencies, can be calculated using Equation 3 (from Dahl, Appendix B), as follows:

$$TS_{(sphere)} = 20 \times log(a)-44 \tag{3}$$

where a = sphere radius (mm).

As with fish, TS of the resonant spheres increases with size (Table 2). At frequencies other than near resonance (> 25% of resonance frequency; Kinsler et al. 1982), the approximate TS of the sphere can be calculated using Equation 1. These calculations are idealized, as neither the effects of fish tissue on sphere resonance nor interference problems due to spectral characteristics of a fish are considered.

Table 1. Theoretical target strengths (TS in dB) of fish in relation to length and frequency (see Equation 2).

Fish		Frequenc	y (kHz)	
length (cm)	10	50	100	500
10	-47.50	-48.40	-48.80	-49.70
30	-38.50	-39.50	-39.80	-40.80
60	-32.90	-33.80	-34.20	-35.10
90	-29.60	-30.50	-30.90	-31.80

Table 2. Target strength, resonant frequency, volume and displacement (buoyancy) of hollow, resonant spheres (air-filled glass spheres; thickness ratio = 0.99).

Sphere radius (mm)	Target strength (dB)	Resonant frequency (kHz)	Sphere volume (mm³)	Sphere displacement (mg)
0.50	-50.00	391.6	0.5	0.5
1.00	-44.00	195.8	4.2	4.2
1.80	-39.00	108.8	24	24.4
2.00	-38.00	97.9	34	34
5.00	-30.00	39.2	520	520

In conjunction with this study, a contract was issued to the University of Washington Applied Physics Laboratory to investigate the theoretical feasibility of a resonating tag for fisheries. The results of the study are presented in Appendix B and are based upon a fixed set of conditions. These conditions were as follows: fish length of 100 mm; two tag sizes, one with a radius of  $\geq 1.0$  mm for a hollow sphere and one with a radius of  $\geq 1.8$  mm for a solid sphere; and frequencies of 30- to 500-kHz).

## **Results and Discussion**

Three critical factors in the development of a resonant tag are 1) the size range of fish to be tagged, 2) the size range of tagged fish to be detected, and 3) the size of the implanted tag. There is a strong relationship between these factors from both biological and acoustical standpoints. The combined reflectivity of the fish and the resonant sphere is equivalent to their combined scattering areas. As a fish increases in size, its target strength also increases (Equation 3), thus eventually masking the acoustic contribution of a resonant tag.

Target strengths (TS) for fish of various lengths (10, 30, and 60 cm), in relation to an 1.8-mm-radius resonant, hollow glass sphere having a shell thickness ratio of 0.99, are shown in Table 3. Table 3 values were calculated for interrogation at tag resonant frequency using Equations 3, 4 (Appendix B), and 5 (from Johannasson and Mitson 1983).

However, at frequencies greater than those where resonance effects are significant, the TS of the sphere will amount to -44 dB (Equation 1) or 5 dB lower than at resonance. Reflectivity at frequencies below resonance would be considerably less.

$$F_{\text{(res)}} = 195.8/a$$
 (4)

where a = sphere radius (mm).

$$TS = 10 \log((\sigma_{(fish)} + \sigma_{(sphere)})/4\pi)$$
 (5)

where  $\sigma_{(fish)}$  = effective scattering area of a fish (m<sup>2</sup>) and  $\sigma_{(sphere)}$  = effective scattering area of an implanted resonant sphere (m<sup>2</sup>).

The theoretical enhancement of fish TS values by a 1.8-mm radius resonant sphere, measured at the resonant frequency, would equal 10.3, 3.3, and 1.1 dB for the 10-, 30-, and 60-cm fish, respectively (Table 3). The 10-dB enhancement of a 10-cm fish would probably be measurable, but the 3-dB enhancement would be near the threshold of discrimination by spectral analyses. It is unlikely that the 1-dB enhancement of the 60-cm-long fish TS would be detectable. A larger resonant sphere would enhance detection in large fish, but if implanted in small fish before they grow to larger size, may also impose an unacceptable biological burden.

An implanted hollow resonant sphere may affect a fish's equilibrium because the sphere will be buoyant. The displacement of a hollow sphere in water (disregarding its weight) can be calculated by the equation:

$$D_{(sphere)} = V_{(sphere)}/r$$
where  $V_{(sphere)} = 4\pi a^3/3$  (cm<sup>3</sup>)
$$D = displacement (g)$$

$$r = density of water (g/cm3)$$
(6)

Displacement of the swim bladder of a 10-cm-long salmon is about 700 mg, while displacement of an 1.8-mm-radius sphere is 24.4 mg, and would represent about 7% of the fish's body weight in fresh water (Shibata 1970, Johannesson and Mitson 1983). Thus, sphere displacement (buoyancy) would amount to approximately 3% of swim bladder displacement. While this would probably not present a significant biological burden, the buoyancy of a larger resonant sphere might (Table 2). For example, a 5.0-mm-radius sphere would displace 520 mg, which would be equivalent to almost 75% of swim bladder displacement in a 10-cm fish.

The discussion of fish marking with resonant spheres has to this point disregarded the effects of environmental factors on detection of tags. The presence of bubbles, environmental noise, and the viewing or aspect angle for interrogation of tagged fish can all limit the ability to detect tagged fish. Bubbles formed by wind, swift current, and waterfalls would interfere with acoustic sound propagation and thus reduce detection efficiency. Bubbles are excellent reflectors that effectively scatter directed sound, often to the extent that a detection system could be disabled. Further, bubbles similar in size to an implanted sphere could resonate and interfere with tag detection. Sites where a resonant-tag interrogation system could be deployed will have to be carefully selected to reduce the effects of bubbles.

Table 3. Theoretical target strength and TS enhancement (dB) of fish due to an implanted sphere having a radius of 1.8 mm and a resonant frequency of 108.8 kHz.

		Target Strength		
Fish length (cm)	Fish (dB)	Sphere (dB)	Combined (dB)	Enhancement (dB)
10.00	-49.00	-39.00	-38.60	10.4
30.00	-39.50	-39.00	-36.20	3.3
60.00	-33.80	-39.00	-32.70	1.1

Strong ambient noise sources will also interfere with resonant tag detection. Spectral components of noise are indistinguishable from those of target fish or resonant tags when they occur in the same frequency range, and their presence could thus mask tag recognition. Potential noise sources include pumps, power turbines, waterfalls, rain, and boats.

The interrogation viewing or aspect angles from which tagged fish could be recognized will be limited by fish morphology. When interrogated from the dorsal aspect, a fish's swim bladder, dorsal musculature, and vertebral column would be aligned in the propagation path between the resonant sphere tag in the coelomic cavity and the interrogating transducer. The result would be that the interrogating acoustic field strength and the echo from the sphere would be significantly attenuated.

A study of acoustical energy transfer through a fish body indicated that this attenuation could block tag detection for aspect angles within at least 45 degrees from dorsal aspect (see "Technical Feasibility of an Acoustic PIT-Tag" in this report). An interrogation transducer would probably have to be deployed such that fish are insonified from side or ventral aspects to obtain reliable detection.

The theoretical work of the University of Washington Applied Physics Laboratory (Appendix B) concluded that hollow spheres of useable size for use in juvenile salmonids could increase their TS by as much as 6 dB, and thus could theoretically be a useful tool for monitoring salmon movement. A limited discussion of sphere material and construction is presented in Appendix B.

Based upon the theoretical calculations of fish/sphere TS and the independent evaluation of the concept of resonant sphere tags by the University of Washington, the proposed detection system appears to be technically feasible and probably could be developed. However, because of the number of factors discussed that will negatively impact system performance, the resonating sphere tag would have very limited application. Thus, we recommend that a resonant sphere tag detection system not be developed for use in the Columbia River Basin at this time.

### **Conclusions and Recommendations**

We concluded that on a theoretical basis, the detection of resonant spheres implanted within fish is feasible but severely limited by several factors. An independent appraisal of the concept by the University of Washington Applied Physics Laboratory confirmed our conclusion. Factors identified as limiting the performance of the system included:

- 1) increasing difficulty to detect tags as fish grow;
- 2) the problem of tag size, which must be considered in terms of biological effect on small fish and loss of detection ability as fish grow larger;
- 3) sensitivity of tag detection, which may be limited by bubbles in the water, ambient noise, and the spectral characteristics of fish; and
- 4) blocking of tag detection by bone, tissue, and swim bladder shading.

Even though development of a resonant sphere detection system is technically feasible, the above limitations so restrict its potential use that we recommend it not be developed for use in the Columbia River Basin at this time.

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# DETRIMENTAL EFFECTS OF ULTRASOUND ON ANIMALS: A LITERATURE REVIEW

#### Introduction

Two types of passive acoustically energized tags were consideration for potential development by NMFS (see "Technical Feasibility of an Acoustic PIT-Tag" and "Technical Feasibility of a Acoustic Resonant Tag" in this report). The first tag, described as an acoustic-passive-integrated transponder (A-PIT), would acquire its operating power and would respond using acoustical (sound) energy. Sound, as used in this paper, refers to any vibration or displacement of water or air particles in response to a pressure wave.

Two separate sound fields would operate the A-PIT tag system: a continuous 50-kHz field would energize the tag, and a 500-kHz field would be used for its response. An energy budget was estimated for the system based on a receiver threshold of 93 dB $\mu$ Pa (decibels referenced to 1.0 micro Pascal) (power level =  $1.3 \times 10^{-7}$  mW/cm²), and a tag detectability range of 10 m. Results suggested the 50-kHz energizing sound-field strength should be 207-213 dB $\mu$ Pa@1 m (330-1330 W/m²) and the 500-kHz response-field strength should be 120-126 dB $\mu$ Pa@1 m (0.67-2.65 mW/m²). Conversions of acoustical pressure to equivalent electrical power are presented for reference in Table 1. Based on these calculations, the minimum energizing field that must strike the tag would be 186-192 dB $\mu$ Pa. Thus, the process of energizing the proposed A-PIT tag will expose target species and other animals to strong, continuous acoustic fields at 50 kHz.

The second tag proposed was a glass sphere that would acoustically resonate at a specific frequency. Such a tag would be detected by spectral analysis of acoustic echo returns from exposing fish to a pulse from a limited portion of a broad frequency band (i.e., 10-500 kHz). The sound-field strength needed for acquisition of sphere echo returns from a 10-m range was estimated at 207-213 dBµPa@1 m from the face of the transducer. This field strength is similar to that proposed for the A-PIT tag, and as such, would likewise expose biota to strong, pulsed acoustic fields.

Based on the above information, a review of the literature was conducted to investigate possible detrimental effects on biota from acoustic energy. Discussions are focused toward the frequency and field strengths required to operate either of the proposed tags.

Table 1. Conversions of acoustical pressure to equivalent electrical power.

Acoustical	
Pressure	Equivalent Electrical power
dB μPa	Watts/m <sup>2</sup>
10.00	$6.667 \times 10^{-18}$
20	$6.667 \times 10^{-17}$
30	$6.667 \times 10^{-16}$
40	$6.667 \times 10^{-15}$
50	$6.667 \times 10^{-14}$
60	$6.667 \times 10^{-13}$
70	$6.667 \times 10^{-12}$
80	$6.667 \times 10^{-11}$
90	$6.667 \times 10^{-10}$
100	$6.667 \times 10^{-09}$
110	$6.667 \times 10^{-08}$
120	$6.667 \times 10^{-07}$
130	$6.667 \times 10^{-06}$
140	$6.667 \times 10^{-05}$
150	$6.667 \times 10^{-04}$
160	$6.667 \times 10^{-03}$
170	$6.667 \times 10^{-02}$
180	$6.667 \times 10^{-01}$
190	$6.667 \times 10^{+00}$
200	$6.667 \times 10^{+01}$
210	$6.667 \times 10^{+02}$
220	$6.667 \times 10^{+03}$
230	$6.667 \times 10^{+04}$
240	$6.667 \times 10^{+05}$

#### **Methods and Materials**

The literature search conducted was limited in scope, concentrating on major papers that best represent the state of current knowledge in the area of acoustics as related to fish, cetaceans, and terrestrial animals. Literature citations were grouped into six discussion areas. The first two areas provided background information on general mechanisms and abilities of fish to sense sound. The third and fourth areas described some effects of acoustic energy on fish behavior and physiological changes that can be caused when fish are exposed to strong acoustic fields. The fifth area discussed hearing sensitivity of cetaceans and the sound fields they produce, and the final area included literature describing detrimental effects of acoustic fields on terrestrial animals.

#### **Results and Discussion**

# Sound Sensing Mechanisms of Fish

Some mechanisms fish use to sense sound are similar to those of terrestrial animals, while others are very different. Fish can sense sound vibrations and water particle velocity and acceleration. Together, these abilities allow some fish to create a three-dimensional sound image of their environment. The overall set of organs and structures used by fish to sense sound is called the octavolateralis system. Detailed descriptions of the morphology, mechanism, and application of the various components of the octavolateralis system have been previously published (Alexander 1962; Buekle 1968; Enger and Anderson 1967; Tavolga 1967; Chapman and Hawkins 1973; Sand and Enger 1973; Fay 1974; Fay and Popper 1974, 1975; Sand 1974, 1981; Hawkins and Johnstone 1978; Myrberg and Spires 1980; Buwalda 1981; Platt and Popper 1981; Buwalda et al. 1983; Popper 1983; Saidel and Popper 1983; Rogers and Cox 1988; Kalmijn 1988, 1989; Platt, Popper and Fay 1989; Bleckman 1993; Enger et al. 1993; Popper and Platt 1993; Carlson 1995).

### **Sound Sensing Abilities of Fish**

Sound-frequency and field-sensing thresholds differ considerably among fish species. For salmonids, sound-frequency sensing thresholds are fairly constant, ranging from <1 Hz to about 150 Hz, but rise steeply for higher frequencies. Near-total loss of sound detection occurs at frequencies >380 Hz (Knudson et al. 1992, Kalmijn 1988). Weber and Schiewe (1976) concluded that the lateral line of salmonids is responsive to

stimulation by frequencies from 1 to 345 Hz, with maximum sensitivity between 10 and 170 Hz. Contrary to the findings of other investigators, Shabalin (1991) reported rainbow trout (*Oncorhynchus mykiss*) can detect sound at frequencies up to 50 kHz at >100 dB  $\mu$ Pa.

Clupeids, such as blueback herring (*Alosa aestivalis*) and shad (*Alosa sapidissima*), are able to detect frequencies ranging from 60 Hz to 150 kHz (Dunning et al. 1992, Pickens 1992, Nestler et al. 1992, Ross et al. 1993). Shabalin (1991) reported golden mullet (*Mugil aratus*) detected sound at frequencies to 4-70 kHz at >92 dBμPa, garfish (*Belone belone*) to 80 kHz at >112 dBμPa, silver carp (*Hypophthalmichthys molitrix*) to 80-95 kHz at >80 dBμPa, and common carp (*Cyprinus carpio*) to 125 kHz at >114-124 dBμmPa. Shabalin found that the transducer he used to generate underwater sound also produced an electromagnetic field (EMF).

After investigating this, he reported that the common carp can detect EMFs from 10 Hz to 160 kHz and can sense changes in EMF levels of 12 dB for field strengths of at least 10<sup>-4</sup> V/cm. Shabalin also reported that all of his study species had some sensitivity to EMFs. Similar findings have not been reported by other investigators. However, it has been shown that cochlear hair cells of some fish and animals are sensitive to electrical current (Brownell and Kachar 1985, Ashmore and Brownell 1986, Jen and Steele 1987).

The above investigations show that the 50-kHz tag-energizing frequency produced by an A-PIT system could be sensed by some fish species, including perhaps salmonids. However, the 500-kHz tag-response frequency is higher than any reported to be detectable by fish. In addition, the sound field produced by the tag (response field) is low (120-126 dB $\mu$ Pa@1 m) and is only present after the tag has been energized. It is not known if a transducer used to produce an A-PIT energizing sound field would also produce an EMF, how strong such a field might be, or how sensitive fish might be to the exposure.

The operating frequency of the proposed resonating sphere tag system would lie between 10 and 500 kHz. As with the A-PIT-tag system, the lower operating frequencies may be audible to fish under certain conditions, since the energizing field strength would be high  $(207-213 \text{ dB}\mu\text{Pa}@1 \text{ m})$ .

#### Fish Avoidance of Sound Fields

There is concern that fish behavior may be modified by sound fields produced by the proposed A-PIT system and the resonant sphere detection system. If fish are repelled or attracted, tag detection data could be biased. This section is a condensed review of sound field levels and frequencies reported to affect fish behavior.

Knudson et al. (1992) reported consistent avoidance by Atlantic salmon (*Salmo salar*) and brown trout (*Salmo trutta*) to a 10-Hz sound field in a pool; no habituation was noted. The authors stated that at 5-10 Hz, particle acceleration values should be at least 0.01 m/s² to elicit an avoidance response in salmonids. Enger et al. (1993) reported that migrating Atlantic salmon smolts consistently avoided a 10-Hz sound field. The effective range was approximately 3 m, within which particle acceleration was greater than 0.01 m/s². However, no behavioral response was noted for a 150-Hz sound field, even when the fish approached to within <10 cm of the sound source.

Particle acceleration at 10 cm from the source was more than 114 dB greater (factor of 500,000) than the measured hearing threshold for Atlantic salmon for frequencies between 5 and 10 Hz (Knudson et al.,1992). VanDerwalker (1967) reported short-range avoidance (0.6 m) by juvenile chinook salmon (*Oncorhynchus tshawytscha*) responding to sound fields at frequencies of 30 to 150 Hz, but awareness dropped off at frequencies higher than 150 Hz. The fish did not appear to habituate to the sound.

Clupeids are sensitive to higher frequencies and to lower sound levels than salmonids. Pickens (1992) and Nestler et al. (1992) reported initial reactions by blueback herring and shad when exposed to sound-field frequencies between 60 and 500 Hz. However, they quickly acclimated to repeated exposure. Both species were attracted to a continuous sound frequency of 80 Hz, but were repelled by frequencies of 100-110 kHz. They were most sensitive to frequencies near 130 kHz, and continuous noise over a band of 100-150 kHz was strongly avoided. Experimentation determined that a 130-kHz sound-field level of 176 dBμPa@1 m caused both species to move at least 7.7 m (158 dBμPa@7.7m).

Effective avoidance to 61 m was obtained when the 130-kHz sound-field level was increased to 183 dB $\mu$ Pa@1 m (reduces to 147 dB $\mu$ Pa@61 m). Dunning et al. (1992) found that blueback herring were repelled to a distance of 60 m by 124.6- and 130-kHz sound fields having a source level of 187 dB $\mu$ Pa@1 m (reduces to 151 dB $\mu$ Pa@60 m).

Reduced response was seen for frequencies below 110 kHz or above 140 kHz. Finally, Carlson (1995) reported that blueback herring showed strong avoidance to a 120-kHz sound field of 200 dB $\mu$ Pa@1 m, with some individuals within 2 m of the source (194 dB $\mu$ Pa@2 m) being stunned or killed.

The reported reactions of fish to various sound-field levels supports the notion that the A-PIT or the resonating sphere tag systems could affect some species of fish when near the sound source. Blueback herring and shad could sense the energizing sound fields of the proposed tags and would very likely react to them. The 50-kHz energizing frequency for the A-PIT tag is below either fish's maximum frequency sensitivity (130 kHz), but some sphere tag frequencies could be within the range of maximum sensitivity for either species.

In addition, the proposed energizing field strength for both tags is very high (207-213 dB $\mu$ Pa@1 m); nearly 70 times higher than levels that repel either shad or blueback herring (176 dB $\mu$ Pa@1 m; Pickens 1992; Nestler et al., 1992). The energizing sound-field levels proposed for both tag systems are greater than those reported to have stunned or killed either species when very near a sound source (Carlson 1995). It is unlikely however, that any fish species would react to a 500-kHz tag response frequency of the A-PIT tag. This frequency is higher than that reported to cause fish avoidance, and the sound-field level is low (120-126 dB $\mu$ Pa@1 m) (Pickens 1992, Nestler et al. 1992, Dunning et al. 1992) and would only be present after a tag was energized.

The 50-kHz A-PIT-tag energizing field or the range of frequencies proposed for resonating sphere tags would unlikely elicit any reaction by salmonids, since both are higher than frequencies generally reported to be detectable by these fish (VanDerwalker 1967, McKinley and Patric 1987, Knudson et al. 1992, Enger et al. 1993). However, Shabalin (1991) reported that rainbow trout (*Oncorhynchus mykiss*) could detect frequencies up to 50 kHz at >100 dB $\mu$ Pa. If this is indeed the case, then some salmonids would be able to detect the proposed tags and thus could respond.

### **Effects of Exposure to Strong Sound Fields**

Exposure to intense sound fields can result in tissue and cellular damage in addition to physiological and neurological trauma to animals. A limited review of pertinent literature is presented below. Intense sound fields, under certain conditions, can produce bubbles, cell wall rupture, and tissue heating. Limited research has been conducted using fish as test subjects, but a wide range of literature describing

experiments using terrestrial animals is available. The results of these studies may be applicable to some fish because of similarities in their inner ear and tissue morphology.

Ter Haar and Daniels (1981) reported that guinea pigs ( $Cavia\ cobaya$ ) exposed for 5 minutes to 750 kHz at an intensity of 150 mW/cm² (213.5 dBµPa) developed 10-µm-diameter bubbles in their blood. They did not detect bubble formation for exposure intensities < 80 mW/cm² (210.8 dBµPa). Other investigators have shown that high-intensity sound fields will damage cellular structures. Dooley et al. (1983) reported that about 60% of rat lymphocytes in suspension were damaged by continuous 10-minute exposures to 500 kHz at 2 W/cm² (224.8 dBµPa). Pulsed ultrasound at the same frequency and equivalent average temporal intensity, but with peak intensity values of 30 W/cm² (236.5 dBµPa), resulted in about the same level of cell damage.

Other than causing direct mechanical damage, exposure to a strong sound field can cause frictional heating in tissue. Martin et al. (1982) found that tails of plattyfish (*Xiphophorous maculatus*), when exposed to 780 kHz at 2.6 W/cm² (225.9 dB $\mu$ Pa), increased in temperature by 2.2 to 3.5°C after 30 seconds. Blood flow increased when the tails were exposed to 780 kHz at 10 mW/cm² (201.8 dB $\mu$ Pa). Maximum blood flow was reached after 5-10 minutes.

No data clearly shows that the energizing sound field of either of the proposed tags would cause tissue heating in fish. Sound-field/frequency combinations reported to cause significant tissue heating were of levels and frequencies higher than those of the proposed tag systems.

The energizing field level (207-213 dB $\mu$ Pa@1 m) of the proposed tags is of equal strength to a 750-kHz field that was shown to cause bubble formation in tissue by cavitation (Ter Haar and Daniels 1981). Cavitation occurs more readily at 750 kHz than at lower frequencies, but fish may approach much nearer to either of the proposed tag system transducers than 1 m. For instance, exposure level at 10 cm from the transducer could be 10 times greater than at 1 m (227-233 dB $\mu$ Pa@10cm) (Kinsler et al. 1982) and may be sufficient to cause bubble formation.

The lateral line of salmonids can be severely disabled by gas bubble disease. Weber and Schiewe (1976) found that steelhead trout exposed to water with a gas pressure of 118% of saturation showed effects of gas bubble disease in the lateral line

within 2-6 hours. Further, progression of bubble formation resulted in increased loss of lateral-line functioning to the point of near-total unresponsiveness. Recovery following return of the fish to equilibrated water required 16-20 hours (Weber and Schiewe 1976). Gas bubble formation in fish caused by exposure to an intense sound field may result in symptoms similar to those of gas bubble disease, or exposure may aggravate a preexisting condition.

Exposure of fish to intense sound fields can result in effects ranging from temporary or permanent loss of sound-detection sensitivity to stunning and death. Hastings (1990) stated that the literature has no reports concerning morphological damage to the lateral-line system caused by intense underwater sound. However, Weber and Schiewe (1976) found that the lateral line of salmonids can be temporarily disabled by high-intensity sound (no sound level reported). Since salmonids have limited ability to detect sound-pressure fields, due to minimal coupling between their inner ear and swim bladder, their ability to avoid potentially damaging conditions is likewise limited.

Other species, so-called "hearing specialists" (eg., goldfish, *Carassius auratus*), are much more sensitive to sound-pressure fields and are susceptible to inner-ear damage by intense sound (Alexander 1962; Buekle 1968; Enger and Anderson 1967; Tavolga 1967; Chapman and Hawkins 1973; Sand and Enger 1973; Fay 1974; Fay and Popper 1974, 1975; Sand 1974, 1981; Hawkins and Johnstone 1978; Myrberg and Spires 1980; Buwalda 1981; Platt and Popper 1981; Buwalda et al. 1983; Popper 1983; Saidel and Popper 1983; Rogers and Cox 1988; Kalmijn 1988, 1989; Platt, Popper and Fay, 1989; Bleckman 1993; Enger et al. 1993; Popper and Platt 1993; Carlson 1995).

Popper and Clark (1976) exposed a hearing-specialist species (e.g., goldfish) to a sound field of 149 dB $\mu$ Pa at frequencies of 300, 500, 800, and 1000 Hz for 4 hours. They measured a temporary shift in hearing thresholds at 500 and 800 Hz immediately after exposure, but all thresholds returned to normal within 24 hours. Hastings et al. (1986, 1987) and Hastings (1990) reported that goldfish were killed, and the otolith organs of other species were severely damaged, by 0.5- to 2-hour exposures to 250- and 500-Hz sound fields of 182-204 dB $\mu$ Pa. At 250 Hz, physical damage to the otolith organs began at a field strength of 189 dB $\mu$ Pa, and at 500 Hz, damage began at 197 dB $\mu$ Pa.

Scanning electron microscopy (SEM) revealed that these sound-level exposures resulted in destruction of hair cell cilia on the maculae of the saccular otolith organ. Some hair cell cilia that appeared (SEM inspection) to be physically undamaged were

also injured. At least a 10-dB increase in the sensitivity threshold of nerve fibers was observed for fish subjected to the lowest field strength exposure (182 dB $\mu$ Pa) for the experiment. Transient stunning also occurred, but its physiological mechanism and threshold values were unknown.

Gourami (*Colisa* sp.) were immobilized by 8- to 30-minute exposures to 150- and 400-Hz sound fields of 98 and 92 dB $\mu$ Pa (Hastings 1990). Hastings also reported that post-exposure behavioral characteristics and conditions of the gouramis ranged from lethargy and loss of equilibrium to internal hemorrhaging. In addition, some apparently unaffected fish sustained physical damage to their inner ear. Hastings (1990) concluded that sound-pressure levels <150 dB dB $\mu$ Pa are not harmful to fish, while exposure to sound levels >180 dB $\mu$ Pa is harmful to many fish. Further, those species with the swim bladder closely associated with the inner ear are most susceptible. Hastings did not report predictions as to damaging sound-level thresholds related to frequency. Enger (1981) found that when cod (*Gadus morhua*) were exposed for 1-5 hours to 50 and 400 Hz at a field strength of 180 dB $\mu$ Pa, ciliary bundles on the saccular maculae of the inner ear were destroyed.

Very strong underwater sound fields that will kill fish can be produced using explosives. Norris and Mohl (1983) state that lethal exposure thresholds for explosives with a short rise-time begin at 229 dB $\mu$ Pa. Similarly, MacLennan and Simmonds (1992) reported lethal thresholds of 229-234 dB $\mu$ Pa@1 m. Fast explosives such as dynamite and TNT had lethal effects for sound levels 5-10 dB lower than for slower-igniting explosives.

The proposed energizing field strength for the two proposed tags (207-213 dB $\mu$ Pa@1 m) falls well within the range of levels reported to damage fish at ranges from the transducer of 10 m or less (Chapman and Hawkins 1973; Popper and Clark 1976; Enger 1981; Hastings et al. 1986, 1987; Hastings 1990; MacLennan and Simmonds 1992; Carlson 1995). In addition, calculation of sound-field levels at 10 cm from the energizing transducer shows that field strength could be as much as 20 dB (10 times) greater than at 1 m (227-233 dB $\mu$ Pa@10cm).

Fish passing within 10 cm of the energizing transducer could be subjected to about the same sound-field strength as considered lethal when produced by explosives (MacLennan and Simmonds 1992, Noris and Mohl 1983). However, the energizing field for the A-PIT tag would be continuous, and may not be as damaging as pulsed sound. In contrast, the high-energy, pulsed energizing field for the resonating sphere tag may be comparable to pulses produced by explosives, and thus may cause damage to animals at short range (e.g., <1 m) (Dancer et al. 1980, Price 1983).

# Hearing Sensitivity of Cetaceans and the Sound Fields they Produce

The proposed A-PIT and resonant sphere systems could potentially be used in areas such as the Columbia River estuary where cetaceans may be exposed to their energizing and response sound fields. Cetaceans present an enigma in that their hearing is very sensitive to a wide range of sound frequencies, but they do not appear to suffer damage from intense sound-field levels produced by themselves or by others in their social groups.

Au and Snyder (1980) conducted an experiment designed to measure the echo-locating ability of bottlenose porpoise (*Tursiops truncates*). They found that this porpoise could produce a 120-kHz sound level of 160 dB $\mu$ Pa@1 m and could detect, with 50% success, a water-filled steel sphere of -41.6 dB target strength at a range of 113 m. The echo level from the sphere at that range amounted to 76.3 dB $\mu$ Pa. Detection success at 100 m (2 dB greater echo level) was 91%.

Other cetaceans are able to produce much higher sound levels. Norris and Mohl (1983) presented convincing evidence that whales are able to stun or debilitate prey from great distances (no distances reported) by emitting sonic beams in the 1-5 kHz band with sound levels of 230 dB $\mu$ Pa@1 m. Similar observations were reported by Hult (1982), but no measurements of source levels were reported. Taylor (1986) also stated that whales are apparently able to stun prey. Most cetaceans can produce and sense high-frequency sound fields (Backus and Shevill 1966, Diercks 1972).

The operating frequency range (30-500kHz) of the proposed tag systems would be audible to some cetaceans, but it is not known if the energizing field strength (207-213 dB $\mu$ Pa@1 m) would present a hazard. Many cetaceans produce echo-location field strengths of nearly the same level, and some produce levels many times as strong (Norris and Mohl 1983). In addition, cetaceans within a social group or pod can apparently withstand exposure to strong sound fields produced by other individuals without suffering injury.

# Damaging Sound Field Level Exposures for Terrestrial Animals

Some aquatic or semi-aquatic animals such as otters or beavers may be exposed to the strong energizing sound field of the proposed A-PIT or resonant sphere tag systems if the systems were deployed in streams or lakes. Little is known about the sensitivity of these animals to the effects of strong sound fields. However, it is well documented that when animal subjects are exposed to excessive noise, temporary and/or permanent reductions in hearing sensitivity can occur.

Continuous exposures to moderate-level noise will cause asymptotic hearing thresholds shifts (ATS) within 18-24 hours. Permanent threshold shifts (PTS) depend upon the level, frequency, and duration of exposure. Below a "critical level" of about 115 dB//20 $\mu$ Pa (141 dB $\mu$ Pa), PTS and hair cell loss in the cochlea are generally related to the total energy received during a continuous exposure. Periodic rest periods inserted in an exposure schedule reduce hearing loss and cochlear damage (Clark 1991).

Several reports have detailed the effects of exposure to strong sound-field levels for short durations (>126 dB $\mu$ Pa) in behaviorally trained animals (Ward and Duval 1971; Lonsbury-Martin and Martin 1981; Borg 1982a,b; Buck et al. 1984). Generally, the noise exposures used in these studies were sufficient to produce PTS after only a few minutes to a few hours. In addition, a correlation was noted between measured permanent hearing loss and the extent and location of damage to sensory cells. Intense, short rise-time sound pulses are more damaging to hearing than equivalent sound-energy exposures to pulses having a more gradual increase in level (Dancer et al. 1980, Price 1983).

The proposed 500-kHz A-PIT-tag response frequency is probably far beyond the hearing range of terrestrial animals, but the sound field (120-126 dB $\mu$ Pa@1 m) approaches levels reported as harmful for long-term exposure (>126 dB $\mu$ Pa, several hours to several days). However, a tag response would be generated only during tag energizing, when an animal would also be subjected to the much stronger 50-kHz A-PIT energizing sound field. This energizing sound field may be audible to some animals. In addition, its field strength (207-213 dB $\mu$ Pa@1 m) is many times greater than the reported threshold for injury (126 dB $\mu$ Pa). If an aquatic mammal were to remain submerged within the A-PIT energizing field for more than a brief period, hearing damage would most likely occur. Similarly, the calculated sound field (207-213 dB $\mu$ Pa@1 m) required to operate the spherical resonating tag creates the same concern for animals.

#### **Conclusion and Recommendations**

A review of literature covering the effect of sound on animals strongly suggested that the sound-field strength of either of the proposed tag-detection systems could, under certain conditions, modify behavior or cause harm to fish, and perhaps also to some aquatic mammals.

Two separate sound fields would be produced by the proposed A-PIT-tag system: the first would be a continuous field at 50 kHz (energizing frequency), and the second at 500 kHz (response frequency). The strengths of the 50- and 500-kHz sound fields will be 207-213 dB $\mu$ Pa@1 m and 120-126 dB $\mu$ Pa@1 m, respectively. The proposed resonating sphere tag system would operate at a frequency between 10 kHz and 500 kHz (pulsed field), and would produce an energizing field strength of 207-213 dB $\mu$ Pa, as measured at a range of 1 m from the transducer. Based on this information, during operation of either of the proposed tag-detection systems, fish and other animals would be exposed to strong sound fields. Some fish and mammal species would be able to sense the 50-kHz acoustic frequency of the A-PIT tag, but its 500-kHz response frequency would probably be beyond detectable limits.

Reported reactions of fish to strong sound-field levels justifies the concern that fish behavior may be modified by the sound fields produced by either tag system. However, salmonids are unlikely to react to the higher frequencies, as most investigators report that frequencies > 400-500 Hz are beyond the upper limit of their sensitivity. Clupeids (blueback herring and shad) and some other fish species would be able to not only sense the 10-kHz or higher sound field, but would likely react to it. The frequency of maximum sensitivity for blueback herring and shad is near 130 kHz. The proposed energizing sound-field level for either tag detection system would be greater than that reported to have stunned or killed blueback herring and shad near a sound source. It is unlikely fish would react to the 500-kHz A-PIT-tag response frequency or to resonant sphere interrogation frequencies greater than about 250 kHz.

The sound fields produced by either of the proposed tag systems would be audible to some cetaceans, but it is not known if the sound would cause them to alter their behavior or if the sound-field levels would present a significant hazard. Many cetaceans produce echo-location field strengths equal to or greater than the interrogation field strength, and individuals within a social group or pod must often be exposed without suffering injury.

Exposure to intense sound fields can result in reduction of sensitivity to sound, damage to the inner ear, gas bubble formation in tissues, tissue heating that can cause cellular damage, stunning, and death to fish or mammals. The proposed energizing field strength for the A-PIT and the resonating sphere tag system fall well within the range of levels reported to cause injury to fish, in some instances even to a range of as much as 10 m from the transducer. Fish exposed at very short range (10 cm or less) could be subjected to about the same sound-field levels as those produced by explosives. If a mammal were to remain submerged near (10 m) a transducer producing an energizing field for more than a very brief period, hearing damage could occur. However, unlike an explosion-produced sound pulse, fish and other animals may be able to avoid the energizing sound field produced by the tag systems through early detection.

Overall, the literature suggests that some animals could detect and be harmed by the sound fields produced by the proposed tag detection systems. This suggestion is based primarily on information derived from the response of test animals held in a confined area during testing. However, the ability of an animal to detect and avoid a potentially damaging sound field would dramatically reduce its risk. On the other hand, if the proposed tag systems were used in situations where animals could not easily escape potentially damaging sound-field levels (i.e., a fish ladder), damage and/or behavior modification could result.

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#### CONCLUSIONS AND RECOMMENDATIONS

The technical feasibility of developing an acoustic passive integrated transponder (A-PIT) tag was investigated. The initial evaluation addressed the attenuation of acoustic energy by the body of a fish. Placing a test transducer in the coelomic cavity of fish produced wide ranges in attenuation values for the frequencies of interest (50 and 500-kHz) in relation to aspect or viewing angle.

The data suggested that a fish's body and air bladder would significantly attenuate the system's acoustic signals at most viewing angles except for those viewed ventrally and to a limited extent laterally. It was also suggested that if the fish was rapidly swimming very near to the transducer that the narrow beam of the system's transducer at that point would not allow sufficient time to energize the tag.

Two separate sound fields would be produced by the proposed A-PIT tag system; a continuous energizing frequency at 50 kHz and a response frequency at 500 kHz. The strengths of the 50 and 500 kHz sound fields were estimated at 207-213 dB $\mu$ Pa@1 m and 120-126 dB $\mu$ Pa@1 m, respectively. Thus, during operation of an A-PIT system, fish and other animals could be exposed to strong sound fields. A literature review showed that the energy field required to energize the A-PIT-tag could under some conditions cause behavior modification and/or damage to some animals.

Technically the A-PIT tag could be developed. This was confirmed by an independent review of the potential system concept. However, based upon the signal attenuation data showing limited operational viewing aspects and the literature review showing a potential risk to fish and other animals from the tag energizing field under certain operating conditions, we recommend that the tag not be developed. The potential advantages of the A-PIT tag system, and the limited number of applications where it could be applied do not warrant the developmental expense or potential risk to animals.

Similarly, upon investigating the technical feasibility of a resonating-sphere tag, it became apparent that the tag could be developed (confirmed by independent review of the concept), but its application would be limited and there would be a risk to animals confined in proximity to the tag energizing system. Factors limiting system performance included its feasibility for small fish only, its diminishing detection ability as a fish grows, the limited codes (resonating frequencies) available because of tag size in relation

to fish size, reduced tag detection ability due to ambient noise, limited viewing aspects because of a fish's physical characteristics, and the acoustic spectral characteristics of fish.

Calculations showed the strength of the acoustic energizing field for the tag could, as with the A-PIT tag, potentially cause behavior modification or damage to fish or other animals under certain conditions. Based on this information we do not recommend the development of a resonating acoustic sphere tag at this time for use in the Columbia River Basin.

The review of literature covering the effect of sound on animals strongly suggested that the interrogation sound field strength of either of the proposed tags could under certain conditions cause harm and or behavior modification to fish and mammals. However, the ability of an animal to detect and avoid a potentially damaging sound field prior to damage taking place reduces this concern. To reduce the above risk, the systems would thus need to be operated in situations that do not confine animals (i.e., use in open water and not in fish ladders).

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# APPENDIX A: Review and Comments on the A-PIT Tag Tests Conducted at the APL/UW Acoustic Calibration Test Barge 7-10 Feb, 1994

# Review and Comments on the A-PIT Tag Tests Conducted at the APL/UW Acoustic Calibration Test Barge 7-10 Feb, 1994

Report to National Marine Fisheries Service

by

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15 October 1994\*

\*Revised 23 February, 1995 at request of NMFS to remove specific reference to a proprietary proposal previously submitted to the government for consideration.

# **Introduction and Summary**

During the period from 1-10 February 1994, an experiment was conducted at the University of Washington APL acoustic test barge located on the Lake Washington Ship canal. The purpose of the experiment was to "determine the effects of the body of a fish on the directivity and the degree of attenuation of acoustical energy transmitted from and received by a transducer implanted in the coelomic cavity of a salmonid."

Two species were used, sockeye salmon and atlantic salmon. The experiment was conducted to gather critical information needed to assess the feasibility of possible future development of an Acoustical Passive Integrated Transponder (A-PIT) tag. One proposed approach for an A-PIT tag concept was provided in response to reference [1]. The purpose of this report is to comment upon the experimental data in the context of that proposed approach.

In summary, we found that the signal attenuation through the fish flesh varied considerably over the span of the experiments. In some cases, little or no attenuation (even some occasional signal reinforcement) was observed. However, in a large number of cases, substantial signal attenuations were measured. The higher attenuation levels (10-20 dB over bare transducer measurements) occurred primarily at overhead and head-on aspects to the fish host.

The case of 20 dB signal attenuation stresses the proposed design. Although there is sufficient design margin with the proposed approach to accommodate these losses, the large attenuations observed in the experimental measurements increase the development risks if the system is required to accommodate these worst case losses under all circumstances. Nevertheless, there *is* enough design margin in the proposed approach to accommodate the 20 dB signal attenuation losses suggested by the experimental data (see section 4 of this report).

Finally, we note that a battery assisted A-PIT tag concept was included in reference as a back-up risk reduction approach in case that development problems for a purely passive tag proved insurmountable. This concept provides a 60 dB design margin on the interrogation link and provides reliable transmit power for the reply link which does not require energy storage at all. In view of the risk reduction alternatives included in the proposal, we consider that the development risks are manageable for an A-PIT tag concept.

# 1.0 Data Description and Preliminary Observations

There were a number of apparent discrepancies with the acoustic data that indicates that measurement and/or calibration errors are present in the data. Most of these are within 3 dB or so, and therefore are not overly significant (at least as long as we do not expect the answers to be more accurate than this.) Some of the discrepancies, however, are much larger (more than 6 dB). In those cases, we tend to believe that the data is faulty and should be disregarded. There are a few in-between cases in which the errors seem to be in the 3-6 dB range. Normally, we would tend to discount the data in this case as well. Unfortunately these discrepancies are in the calibration plots on the bare transducer. Since these plots have been used as ground truth in all subsequent comparisons, these discrepancies are somewhat troublesome. Some discussion of this problem is provided below in the section on "Bare Transducer Calibration Plots."

### **Index to Data Sets**

In making comparisons, the following table is useful in identifying the appropriate data sets. These references apply to the master data collection log provided by NMFS which indicates what the experimental conditions were for each measurement set. This table allows us to cross reference the various plots by measurement set #, file #, or plot # which are the three ways the sets are referenced in the data log, EXCEL plots, and data plots respectively.

Meas.	File	Plot	Meas	Xducer	Bladder	Fish	Group
Set	No.	No.	Axis	Config	Cond	Length	
1	XDR-H-R.50	1944	Pitch	Bare	NA	NA	1
2	XDR-H-R.500	4188	Pitch	Bare	NA	NA	1
3	XDR-H-T.500	1947	Pitch	Bare	NA	NA	1
4	XDR-V-T.500	1948	Roll	Bare	NA	NA	2
5	XDR-V-R.500	4189	Roll	Bare	NA	NA	2
6	XDR-V-R.50*	4190	Roll	Bare	NA	NA	2
7	F-1-V-R.50	4191	Roll	External	Def (?)	24.8	3
8	FlA-V-R.50	4192	Roll	External	Def (?)	24.8	3
9	F1B-V-R.50	4193	Roll	External	Def (?)	24.8	3
10	F1C-V-R.500	4194	Roll	External	Inflated	24.8	3
11	F1D-V-T.500	1949	Roll	External	Inflated	24.8	3
12	F1E-V-T.500	1950	Roll	External	Inflated	24.8	3
13	F1F-V-R-500	4195	Roll	External	Inflated	24.8	3
14	F1G-V-R.50	4196	Roll	External	Inflated	24.8	3
15	F2A-V-R.50	4197	Roll	Internal	Inflated	26.7	4
16	F2B-V-R.500	4198	Roll	Internal	Inflated	26.7	4
17	F2C-V-T.500	1951	Roll	Internal	Inflated	26.7	4
18	F2D-V-T.500	1952	Roll	Internal	Pt Def	26.7	4
19	F2E-V-R.500	4199	Roll	Internal	Pt Def	26.7	4
20	F2F-V-R.50	4200	Roll	Internal	Pt Def	26.7	4

21	F3A-N-R.50	4201	Yaw	Internal	Inflated	26.7	5
22	F3B-N-R.500	4202	Yaw	Internal	Inflated	26.7	5
23	F3C-N-T.500	1954	Yaw	Internal	Inflated	26.7	5
24	F3D-N-T.500	1955	Yaw	Internal	Deflated	26.7	5
25	F3E-N-R.500	4203	Yaw	Internal	Deflated	26.7	5
26	F3F-N-R.50	4204	Yaw	Internal	Deflated	26.7	5
27	F3G-H-R.50	4205	Pitch	Internal	Inflated	26.7	6
28	F3H-H-R.500	4206	Pitch	Internal	Inflated	26.7	6
29	F3I-H- T.500	1956	Pitch	Internal	Inflated	26.7	6
30	F3J-H- T.500	1957	Pitch	Internal	Pt Def	26.7	6
31	F3K-H-R.500	4207	Pitch	Internal	Pt Def	26.7	6
32	F3L-H-R.50	4208	Pitch	Internal	Pt Def	26.7	6
33	F3M-H-R.50	4209	Pitch	Internal	Deflated	26.7	6
34	F3N-H-R.500	4210	Pitch	Internal	Deflated	26.7	6
35	F3O-H-T.500	1958	Pitch	Internal	Deflated	26.7	6
36			N	Missing Data	Set		
37	F4A-V-T.500	1960	Roll	External	Inflated	54.6	7
38	F4B-V-R.500	4211	Roll	External	Inflated	54.6	7
38a	F4C-V-R.50	4212	Roll	External	Inflated	54.6	7
39	F4D-V-R.50	4213	Roll	Internal	Inflated	54.6	8
40	F4E-V-R.500	4214	Roll	Internal	Inflated	54.6	8
41	F4F-V-T.500	1961	Roll	Internal	Inflated	54.6	8
42	F4G-V-T.500	196	Roll	Internal	Deflated	54.6	8
43	F4H-V-R.500	4215	Roll	Internal	Deflated	54.6	8
44	F41-V-R.50	4216	Roll	Internal	Deflated	54.6	8
45	F5A-V-R.50	4218	Roll	Internal	Inf(?)	61.0	9
46	F5B-V-R.500	4220	Roll	Internal	Inf(?)	61.0	9
47	F5C-V-T.500	1963	Roll	Internal	Inf(?)	61.0	9
48	F5D-V-T.500	4221	Roll	Internal	Inf(?)	61.0	9
49	F5E-V-R.500	4222	Roll	Internal	Inf (?)	61.0	9
50	F5F-V-R.50	4223	Roll	Internal	Inf (?)	61.0	9
51	F5G-V-R.50	4224	Roll	Internal	Inf (?)	61.0	9
52	F5H-V-R.50	4225	Roll	Internal	Deflated	61.0	9
53	F5I-V-R.500	4226	Roll	Internal	Deflated	61.0	9
54	F5J-V-T.500	1964	Roll	Internal	Deflated	61.0	9
55	F5K-N-T.500	1965	Yaw	Internal	Inflated	61.0	10
56	F5L-N-R.500	4227	Yaw	Internal	Inflated	61.0	10
57	F5M-N-R.50	4228	Yaw	Internal	Inflated	61.0	10
58	F5N-N-R.50	4229	Yaw	Internal	Deflated	61.0	10
59	F5O-N-R.500	4230	Yaw	Internal	Deflated	61.0	10
60	F5P-N-T.500	1966	Yaw	Internal	Deflated	61.0	10
61	F5Q-H-T.500	1967	Pitch	Internal	Deflated	61.0	11
62	F5R-H-R.500	4231	Pitch	Internal	Deflated	61.0	11
63	F5S-H-R.50	4232	Pitch	Internal	Deflated	61.0	11

Note: The data file indicated for this set in the NMFS master data log was XDR-V-R.500 We believe this was in error.

The groupings in the last column are intended to group measurement sets which are similar with respect to Measurement Axis, Transducer Configuration, and Fish Size. The conditions for inflation/deflation of the air bladder are suspect in many cases because of the difficulty in determining the exact condition of this organ. The table does not attempt to indicate exact transducer placements within or on the fish for internal and external transducer configurations. Some additional details are available in the NMFS data log. See the individual writeups in the NMFS data log for additional comments and details on the conditions of the air bladder, on transducer placement within the fish body cavity, and other details of the measurement sets which we could not summarize in tabular fashion.

#### 2.0 General Remarks

This section provides some general observations as to the apparent validity of the measurement sets. The bare transducer calibration plots (measurements sets 1-6) show Some discrepancies (approximately 6 dB) which are troublesome. Furthermore, they are not consistent with some of the early bare transducer measurements taken by APL/UW prior to beginning the actual tests; i.e. those indicated as "Test Transducer Measurements" and labeled with the acronym NMFS on the plots. Specifically these include plot numbers (1940, 1942, 1943, 4182, 4183, 4184, and 4185). Plot number 1941 appears to be missing in our data set. There were no details on the conditions under which these earlier measurements were taken. We believe that they were not taken in the same support frame as were all of the other measurements, but we're not certain. Hence, these measurements have not been included in our index to the data sets. The plots were, however, included in the data package forwarded by NMFS and we have included these in our comments below.

# **Bare-Transducer Calibration Plots (Measurement Sets 1-6)**

Measurement sets 1 through 6 represent calibration plots for the bare transducer. Data sets 1 and 6 compare the 50 kHz receive patterns about the transducer pitch and roll axes respectively. The roll-axis is along the longitudinal axis of the cylindrical transducer and the pitch axis is at right angles. We would expect the roll pattern to be omnidirectional. This is, in fact, the case. The roll-pattern is omnidirectional within a dB or so. We would expect the two patterns to have the same value at the 90 and 270 degree points; i.e., the points at which the roll and pitch axis measurements present the same transducer orientation to the measurement setup. We see that this is, in fact, the case (at least within a couple of dB or so).

Measurement sets 2 and 5 show the same receiver pattern comparisons but at 500 kHz rather than 50 kHz. These comparisons are not nearly as good as the 50 kHz results. The roll-axis pattern is not very omnidirectional. It has a 5 dB dip at about a 120 degree aspect. Furthermore it does not agree with the "NMFS Test Transducer Measurements" taken by APL/UW prior to the beginning of the test series. These earlier bare-transducer plots show a maximum-to-minimum roll-axis variation of only 2.5 dB by comparison (e.g., see plot 4183 as compared to measurement set #5). A second problem is indicated by the pitch axis pattern (measurement set #2) which shows an approximate 10 degree orientation registration error. This is probably indicative of a misalignment within the support frame. The earlier NMFS measurement (plot 4184) dos not show a similar misalignment. If there is a possibility of this much misalignment in the data measurements using the support frame, the difference patterns between the in-fish and bare-transducer measurements are probably misleading. Misalignments with respect to a null in the bare-transducer pattern could show an apparent gain with respect to the "in-fish" measurement when compared on the basis of difference patterns. This is apparent in several of the difference measurement sets (e.g., sets 22-25, 28-31, 34-35 and 56 are prominent examples). The apparent gains (as much as 15-20 dB) shown in the difference patterns for these sets are probably not real gains, but are very likely an artifact of orientation misalignments between pattern nulls in the two sets of measurements. Finally, we note that the roll-axis pattern does not agree with the pitch-axis pattern at the 90 and 270 degree orientations, as it did at 50 kHz. Even with axis misalignments, we would expect that the peaks in the pitch and roll patterns should agree, since both the pitch and roll axis measurements must cross the transducer's "equator" twice in each rotation. The peaks differ by about 6 dB, indicating a 6 dB discrepancy in these two data sets. This is somewhat disturbing, since these are the bare-transducer calibration plots used for all of the latter comparisons (i.e., in-fish vs bare-transducer comparisons).

Measurement sets 3 and 4 show the same comparisons at 500 kHz but for the transmit patterns instead. These patterns show virtually identical characteristics. This at least shows that transmit/receive reciprocity comparisons are probably valid. Unfortunately it sheds no further light on the discrepancies indicated above. It is possible that the support frame is affecting the patterns. It is also possible that the roll axis measurements were not made at an angle which is precisely perpendicular the transducers longitudinal (roll) axis. This would be a possible explanation for the discrepancies if the roll-axis measurements were off-perpendicular by a constant 15-20 degrees or so; e.g., if the source and receive hydrophones were at different depths by about 1 foot. We don't think that this was a likely event. Furthermore, these same characteristics were not seen in the earlier APL/UW measurements. It would be interesting to know how these earlier measurements were taken; i.e., with or without a frame. If no frame was used, how was

the transducer supported? In any case, we apparently must accept the possibility that the calibration pattern measurements used for comparisons may be in error by as much as 6 dB (a somewhat troublesome prospect).

# **Reciprocity Comparisons**

One of the ways to check the calibration is to compare the transmit and receive data for the in-fish measurements with the corresponding results for the bare transducer. We performed eyeball comparisons with all of the available receive/transmit data sets. The approximate reciprocity discrepancies are indicated below. These are only approximate, and represent only an overall eyeball judgement of the transmit and receive pattern relative differences.

<b>D</b>	α .	ъ.	ъ.	• .
I )ata	Set	Pairs	Recu	procity
Dutt	$\mathcal{L}_{\mathcal{L}}$	1 uns	10001	procity

Data Set I all's	recipiocity
	Discrepancy(dB)
10/11	0  dB
12/13	0
16/17	0
18/19	0
22/23	0
24/25	3
28/29	2
30/31	2
34/35	3
37/38	0*
40/41	2
42/43	1
46/47	3
48/49	2
53/54	1
55/56	2
59/60	3
61/62	3

<sup>\*</sup> Note: There initially appeared to be a 12 dB discrepancy in the EXCEL plot comparisons for this set (37/38). Fortunately this was due to the use of the wrong calibration pattern for data set No.38 in the EXCEL plots. When the correct pattern is used, the results are in very close agreement.

These discrepancies have a mean value of 1.5 dB with a standard deviation of 1.25 dB. We consider these to be reasonable comparisons and would say that the reciprocity comparisons are within the allowable measurement errors for the indicated test setup.

### 3.0 Observations on Similar Measurement Sets

In the following subsections, we have grouped measurements into similarity categories which have the same frequency (either 50 or 500 kHz), measurement axis (pitch, roll or yaw), and transducer configuration (internal or external). These are groupings for which the pattern measurements relative to those of the bare transducer would be expected to be similar. We did not group according to air bladder characteristics or to fish size, since we did not see a consistent correlation with either of these variables. We did not differentiate between transmit and receive patterns because of the close agreement with the reciprocity comparisons discussed above. The measurement sets which fall into the same similarity categories by this criteria, listed by the group index (last column) in our master index, are:

Group	Similarity Category
1	Bare Transducer Pitch-Axis Measurements
2	Bare Transducer Roll-Axis Measurements
3, 7	External Transducer Roll-Axis Measurements
4, 8, 9	Internal Transducer Roll-Axis Measurements
5, 10	Internal Transducer Yaw-Axis Measurements
6, 11	Internal Transducer Pitch-Axis Measurements

Groups 1 and 2 represent the bare transducer only, and are used as the calibration measurements (ground truth) in the absence of the fish structure. The last four similarity groups are discussed individually below for the 50 kHz and 500 kHz measurement sets respectively.

#### External Transducer Roll-Axis Measurements at 50 kHz

The measurement sets which fall into this general category are: 7, 8, 9, 14, and 38a. With the exception of set 38a, the measurements are very similar and do not show drastic or dramatic signal degradation relative to the bare transducer measurement (set # 6). The maximum degradation is about 10 dB and degradations of this magnitude only occur over relatively small azimuth sectors. The azimuth sectors at which the fish seems to attenuate the signal are not consistent among the plots. We did not know on which side of the fish the transducer was mounted for these measurements. There does not seem

to be much correlation with air bladder inflation. A dramatic change is seen in measurement set # 38a. This plot shows a large reduction (10 to 30 dB) in signal receive sensitivity over all azimuths. This particular measurement seems to be at odds with all of the others. We tend to discount this particular measurement set as being unreasonable. If this is a valid measurement, we do not understand the mechanism by which this large signal degradation arises.

#### **Internal Transducer Roll-Axis Measurements at 50 kHz**

The measurement sets which fall into this general category are: 15, 20, 39, 44, 45, 50, 51, and 52. Once again, these measurements are fairly consistent with one notable exception. They generally show that the signal is significantly degraded (10 to 30 dB) on one side, typically when the air bladder is between the transmitter and receiver. The response on the other side is not significantly reduced and in some cases the response is slightly increased. Once again, we have a dramatic exception to these general results, namely set # 15. This plot shows a large reduction (15 to 30 dB) in signal receive sensitivity over all azimuths. Again, this seems to be a flyer. We have no explanation as to the mechanism that would result in this kind of signal absorption at all azimuths; particularly when all other measurements show no indication of this characteristic. We tend not to believe set # 15. No particular correlation with signal degradation and the degree of air bladder inflation is noted.

# Internal Transducer Pitch-Axis Measurements at 50 kHz

The measurement sets which fall into this general category are: 27, 32, 33, and 63. They show a large signal degradation (on the order of 20 dB) from above and directly head-on to the fish. The signal reduction below the fish is non-existent for two of the sets (33 and 63) and is moderately significant (5-10 dB) in sets (27 and 32). This data is consistent with the roll-axis measurements discussed above. The largest reductions occurred with an inflated air bladder (one case only).

### Internal Transducer Yaw-Axis Measurements at 50 kHz

The measurement sets which fall into this general category are: 21, 26, 57, and 58. They show a large signal degradation (on the order of 20 dB) for a directly head-on aspect to the fish. The signal reduction to the sides of the fish vary from as much as 10 dB in set # 21 to a signal reinforcement of as much as 5 dB in sets # 57 and #58. Once again these measurements are consistent with the roll and pitch-axis measurements discussed above. No particular correlation with signal degradation and the degree of air bladder inflation is noted.

#### External Transducer Roll-Axis Measurements at 500 kHz

The measurement sets which fall into this general category are: 10, 11, 12, 13, 37, and 38. The first four sets are consistent and show very little difference in transducer response with and without the fish present. The patterns show considerable fine scalloping presumably due to coherent combinations of reflections from the internal fish structure. There seems to be no correlation with fish bladder inflation and hence the internal reflections do not appear to be dominated by the air bladder. Measurement sets #37 and #38 show quite different results with average degradations of 5 to 10 dB or more. We have no explanation for the apparent difference of these results with the previous ones unless it has to do with the detailed placement of the transducer within the fish cavity. A more careful look at these results with respect to detailed transducer placement may be warranted.

### Internal Transducer Roll-Axis Measurements at 500 kHz

The measurement sets which fall into this general category are: 16, 17, 18, 19, 40, 41, 42, 43, 46, 47, 48, 49, 53, and 54. The first four plots show a very consistent pattern with significant signal degradation (10-20 dB) when the fish is viewed from above. There appears no real difference with air bladder inflation other than a slight reduction of the degradation from above when the bladder is deflated. The remaining measurements are more variable and show scattered regions of signal degradation and reinforcement. No correlation with air bladder inflation is noted.

#### Internal Transducer Pitch-Axis Measurements at 500 kHz

The measurement sets which fall into this general category are: 28, 29, 30, 31, 34, 35, 61, and 62. These plots all show significant signal degradation at most aspects, particularly from above the fish (typically 10-20 dB). They also show significant degradation at tail-on aspects. Signal degradation below the fish is not as pronounced but is still significant in some cases and varies from 5-10 dB. No correlation with air bladder inflation is noted

#### Internal Transducer Yaw-Axis Measurements at 500 kHz

The measurement sets which fall into this general category are: 22, 23, 24, 25, 55, 56, 59, and 60. These plots show a relatively consistent 5-10 dB signal degradation directly to the sides of the fish. This seems a little strange since there should be no strong absorption or scattering mechanisms in these directions. Furthermore, this is not what would be expected from the roll axis measurements at this same aspect. No correlation with air bladder inflation is noted.

# 4.0 Implications for the A-PIT Tag Concept

In this section we briefly discuss the implications of these measurement data relative to the acoustic passive integrated transponder (A-PIT) tag concept which was a proposal presented in response to reference [1]. That concept was a completely passive miniaturized acoustic tag capable of being activated and energized by an acoustic interrogation signal operating at 50 kHz. The tag replies with a unique encoded response at 500 kHz for fish identification and censusing purposes. The baseline proposal was a completely passive tag using a single transducer. Various alternatives were presented as backup approaches as part of the overall risk mitigation plan. These included a dual transducer concept using PVDF for improved receive sensitivity as well as battery-powered and battery-assisted options. Components were identified and preliminary evaluations presented for each option to ensure a low-risk alternative to the baseline approach.

The two primary problems to overcome with the A-PIT tag approach were:

- 1) Providing a sufficiently intense interrogation signal at 50 kHz to energize the tag, and
- 2) Providing a sufficiently strong reply signal at 500 kHz to propagate a signal back to the census station receive array.

We'll consider these two problems separately below. In the following discussions, we'll take the measurements at face value and assume them to be correct. We'll ignore the 6 dB discrepancies noted previously in the bare transducer measurements at 500 kHz.

## The 50 kHz Interrogation Problem

The measurement data indicate that the tag receive response at 50 kHz is relatively highly attenuated (up to about 20 dB or so) at overhead and head-on aspects to the fish. At most other aspect angles, the signal shows much less attenuation or no attenuation at all. In some cases the response is enhanced by the fish presence, but seldom by more than about 3 dB. Nevertheless, a problem still exists in some situations wherein the tag must be energized at overhead or head-on aspects.

The proposal recognized the interrogation link as the critical portion of the system. For this reason a number of alternative schemes were considered to ensure the adequacy of the design in this respect. One of the principal features of the proposed approach was to include multiple interrogation arrays to ensure that the fish would be ensonified at multiple aspects. A properly designed interrogation setup with multiple interrogation arrays would not allow fish passage without providing an interrogation signal at a favorable aspect. The measurement data set supplied by these experiments provides a suitable database for planning an appropriate interrogation setup geometry.

If the multiple interrogation array approach is impractical or unacceptable, the use of a dual transducer concept could be considered. The proposed design opted to use a single PZT transducer ceramic for both transmit and receive. It proposed a system with sufficient design margin to support proper tag operation using the PZT ceramic for both functions. PZT is preferred for transmit but not for receive. An alternative is to transmit on the PZT ceramic and receive on a separate PVDF ceramic which has a receive sensitivity approximately 20 dB higher than PZT. This approach provides 20 dB additional receive sensitivity to compensate the 20 dB fish flesh attenuation losses indicated by the measurement data. If this approach is considered, additional analysis is required to ensure that the energy transfer relationships and transducer impedance characteristics adequately support completely passive tag operation for this configuration.

Finally, battery-assisted operation was considered in which the interrogation signal serves only to trigger the tag. The power necessary to respond would be provided by an internal Lithium Carbon Mono-Fluoride battery, the commercial version of which has a 10 year shelf life with less than 1 percent per year dissipation. Battery dimensions are 2.2 mm by 10 mm. With this approach, the 50 kHz interrogation signal only needs to be sufficient to trigger the tag. The design margin in this case is in excess of 60 dB. This completely offsets the 20 dB fish flesh

attenuation losses indicated by the measurement data and still provides more than a 40 dB design margin on this link. Although this approach does not represent a completely passive tag, it does provide a very attractive alternative to achieve transponder mode operation in a tag which will likely outlast the lifetime of the fish host.

## The 500 kHz Reply Problem

The measurement data indicate that the tag transmit response at 500 kHz can also be attenuated (up to about 20 dB or so) at overhead aspects. This is not always the case, but appears to occur often enough that 20 dB losses cannot be considered completely atypical. Fortunately, we have considerable design margin on this link. The PZT ceramic transducer is capable of transmitting a 143 dB source level. (Note that the APL/UW transducer provided 145-147 dB source level at 500 kHz as shown in Test Transducer Measurement plot #1942). The proposal, however, showed that the required source level is only 123 dB for a 10 meter range. This figure includes a conservative 10 dB design margin as well. Increasing the tag source level another 10 dB (to 133 dB) provides the additional margin necessary to overcome the apparent 20 dB fish flesh attenuation losses indicated by the measurement data. This still leaves us with a 10 dB design margin since the maximum transducer source level capability is 143 dB. The increase in transmit power necessary to raise the transmit source level to 133 dB is not prohibitive. This is because the bulk of the required tag power goes into the electronics rather than into acoustic energy in the water. An increase in source level of 10 dB requires only a 2.2 dB increase in overall tag power requirements. Hence this approach is very practical.

A final alternative which works on both links is to reduce the maximum operating range (R). Signal strength increases inversely as  $20 \log (R) + \alpha R$ . Hence halving the range increases the signal level by more than 6 dB. This may not be a desirable method to recover the entire 20 dB. Nevertheless, it may be reasonable to use this approach to recover a portion of the loss, especially in the overhead (or depth) dimension, which is the primary direction in which fish flesh attenuation seems to be a problem. A full 10 meter range in this dimension may not be necessary.

## Conclusion

Even though the measurement data indicate that considerable fish flesh attenuation may exist for both 50 kHz and 500 kHz transmissions, the proposed A-PIT Tag concept is still viable. The proposed approach provided very conservative design margins and a number of alternative approaches as part of the risk mitigation plan. We

see that a number of alternatives are available to offset the apparent signal losses that may be present on the 50 kHz interrogation link and that sufficient design margin is available to offset apparent signal losses on the 500 kHz reply link as well. These may be used individually or in combination to provide sufficient margin to offset these losses. Even though the measurement data indicate that significant losses may exist for internally placed acoustic tags at certain aspects, these results do not imply that the proposed A-PIT tag concept is infeasible.

## References

[1] Federal Solicitation Number 52ABNF300018 ("Development of Acoustic PIT Tag System"), NOAA, Oct 30, 1992

# APPENDIX B: A Study on the Feasibility or Using Small Spheres to Enhance Fish Target Strength

# A Study on the Feasibility or Using Small Spheres to Enhance Fish Target Strength

by Peter H. Dahl

Letter Report to NMFS, September 1994 Applied Physics Laboratory University of Washington Seattle, Washington 98105

## 1. Introduction

This report summarizes findings of a study to address the feasibility of placing small spheres inside salmon smolts, in order for the fish to be identified acoustically. The basic idea is that a fish which has implanted within it a sphere (referred to as tagged fish), will exhibit a significantly higher acoustic target strength (*TS*) than untagged fish. This *TS* enhancement would occur only at select acoustic frequencies known to excite the acoustic resonances of the sphere. Different spheres sizes and therefore different interrogation frequencies could then be used to distinguish different fish populations.

Spheres are considered because their scattering properties are independent of the direction of incoming acoustic radiation. The sphere acts as a passive transponder: scattering acoustic radiation only when excited by its resonance frequency. Implanted transducer material would not work for this purpose, since good transducer radiation properties do not equate to good passive scattering properties.

The scope of this study is limited to feasibility only. Biological issues (such as fish mortality when tagged) and manufacturing issues (such as costs and availability of spheres) are not addressed.

The study concludes that spherical shells made of glass with radius  $a \ge 1$  mm, or solid spheres made of polystyrene with radius  $a \ge 1.8$  mm, can increase the *TS* of salmon smolts by 6 dB or more. Therefore this method of tagging fish is a potentially useful technique. Recommendations for the next stage of this project are also made.

## 2. Target Strength of Salmon Smolts

We assume the salmon smolts are nominally 100 mm in length. For frequencies between 30 and 500 kHz the fish length (L) to acoustic wavelength ( $\lambda$ ) ratio varies between 2 and about 30. A simple model for fish target strength averaged over all aspects and which spans this  $L\lambda$  ratio is (Love, 1977)

$$TS = 10\log(L^{1.87}\lambda^{0.13}) - 27.7\tag{1}$$

where L and  $\lambda$  are in meters, and TS is in dB. Using L = 0.1 m, Eq. [1] gives  $TS \approx$  -49 dB, with very little frequency dependence over the frequency range of interest. We thus adopt -49 dB as a nominal value for the TS of a 100 mm salmon smolt. We also have confidence in this estimate as it applies to salmonids since actual measurements of salmonid TS at 420 kHz (Dahl and Mathisen, 1983) are predicted to within  $\pm 2$  dB using Eq. [1].

## 3. Enhanced Fish Target Strength Using Implanted Sphere

Assuming incoherent acoustic scattering, then a fish with  $TS \approx -49$  dB which has within it implanted a small sphere of TS = -44 dB will show an increase in target strength about 6 dB. We are seeking at least a 6 dB enhancement in fish target strength, thus small spheres with a  $TS \ge -44$  dB are of interest. Note that more reliable discrimination between tagged and untagged fish would occur if the TS difference was 10 dB or more. To realize a 10 dB difference in TS the sphere TS must be -39 dB.

## **4. Target Strength or Small Spheres**

In this section results of a parameter study on the target strength of small candidate spheres are presented. Both solid spheres and hollow spherical shells are considered. The spherical shell calculations were made by Dr. Steve Kargl of the Applied Physics Laboratory, based on the work contained in Kargl and Marston (1991). Using the notation from this reference, the sphere target strength is defined as

$$TS = 10\log\frac{|F|^2 a^2}{4}$$
 (2)

where |F| is the magnitude of the complex scattering amplitude (in steady-state) of the form function, in the backscattered direction, and a is the sphere radius in meters. Large values of |F|, and therefore TS, exist with certain combinations of frequencies and sphere radii associated with acoustic resonances of the sphere.

A critical parameter is the radius *a* of the sphere (in m if used in Eq. [2]). For example, PIT tags of a maximum dimension equal to 2 mm have been implanted within salmon smolts with fish remaining viable. Other parameters are the sphere material, and in the case of hollow spherical shells, the shell thickness. The shell thickness is defined by the ratio of the inner radius to outer radius. For example, if the radius *a* equals 1 mm, and thickness ratio equals 0.9, then the shell thickness is 100 µm. The hollow spherical shells offer some advantage because they will in general display a large monopole resonance, like an air bubble. The closer the thickness ratio is to unity (or the thinner shell is), the more the sphere behaves like an acoustic bubble.

Three candidate materials were studied: stainless steel, glass, and polystyrene. These were chosen because of their availability, cost, and that these materials are all likely to be biologically inert. Figs. 1-4 show |F| vs ka for glass and polystyrene material spheres. Stainless steel spheres did not offer any advantage over spheres made of glass or polystyrene, and was not considered further. Note that k is acoustic wavenumber which we convert to acoustic frequency assuming a sound speed of 1500 m/s.

Results for a glass spherical shell with thickness ratio = 0.99 (Fig. 3) show a maximum |F| = 12.42 at ka = 0.82, which upon using Eq. [2] translates to

$$TS = -44 + 20\log a_{mm} \tag{3}$$

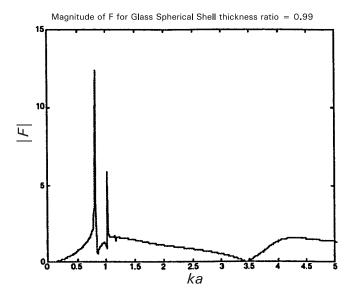


Figure 1: Magnitude of form function F for glass spherical shell vs ka. Material density = 3600, longitudinal speed = 5260, transverse speed = 2960 (MKS).

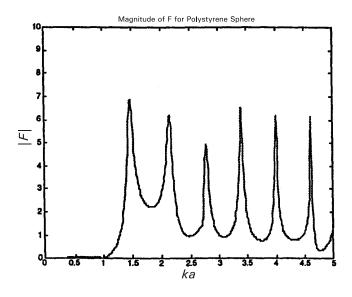


Figure 2: Magnitude of form function F for solid polystyrene sphere vs ka. Material density = 1056, longitudinal speed = 2350, transverse speed = 1120 (MKS).

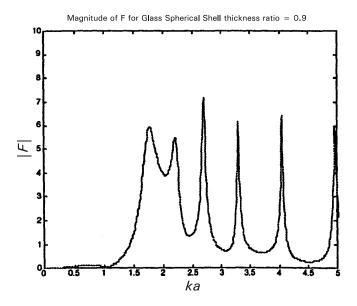


Figure 3: Magnitude of form function F for glass spherical shell vs ka. Material density = 3600, longitudinal speed = 5260, transverse speed = 2960 (MKS).

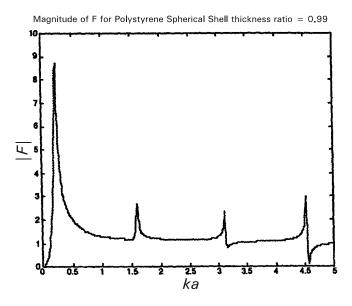


Figure 4: Magnitude of form function: F for polystyrene spherical shell vs ka. Material density = 1056, longitudinal speed = 2350, transverse speed = 1120 (MKS).

in dB, where  $a_{mm}$  is spherical radius is now expressed in mm. For particular radius,  $a_{mm}$  the maximum TS will occur at a frequency f (in kHz) equal to  $195.8/a_{mm}$ . For example, with sphere radius of 1 mm, the TS is -44 dB at a frequency of 195.8 kHz. Increasing the radius to 1.5 mm gives a TS of -40 dB at a frequency of 130.5 kHz. The -40 dB TS value is certainly the more desired scattering level, but it comes at a cost of the spherical radius slightly exceeding the maximum tolerable value as suggested by experience with the PIT tags.

The solid polystyrene sphere data (Fig. 2) is of special interest because one may imagine this type of sphere to be extremely easy to manufacture while also maintaining consistency. The resonance peak of |F| = 6.91 at ka = 1.47, will produce a TS that exceeds -44 dB only for  $a_{mm} > 1.8$ . Although this radius exceeds the maximum tolerable radius, a more exhaustive search of materials may point to other materials similar to polystyrene which produce sufficiently high scattering with smaller spheres.

Another possible candidate is a glass sphere with thicker shell ratio = 0.9 (Fig. 3). Three resonant peaks for ka < 3 may be exploited. The thicker glass shell may also be easier to manufacture. Finally, spherical shells made of polystyrene (Fig. 4) do not seem to offer a significant advantage over solid polystyrene.

In practice, the observed TS will be the result of integration in the frequency-domain of S, the incident signal spectrum; F, as defined above; and H, the receiver frequency response function (Foote, 1983). The backscattering intensity I is thus proportional to

$$I \propto \int_0^\infty |SFH|^2 df \tag{4}$$

Our computations using the results in Figs. 1-4 are based on a narrow band pulse that approximates a single frequency. Since a typical fisheries sonar operates with a relatively narrow bandwidth H  $\approx$ 2-kHz, then I is approximately proportional to  $|F|^2$  evaluated at the center frequency. In the follow-on study (outlined below) the exact integral in Eq. [4] should be evaluated to obtain a more precise value of the sphere TS.

## 5. Summary and Recommendations for Next Stage

This study has demonstrated that implanted small spheres inside salmon smolts can increase the TS of the fish by 6 or more dB. Hollow spherical shells seem the most promising at this stage, e.g., a hollow glass sphere of thickness ratio = 0.99 giving a monopole resonance at ka = 0.83. But there is the possibility that solid polystyrene (or other plastic-like material) spheres may be also be useful, particularly if the maximum allowable sphere radius can be increased. The increased TS, occurring only at a frequency corresponding to the resonances of the sphere, may be used to distinguish different populations of smolts.

We recommend that a more detailed parameter study combined with experimental test be undertaken. The parameter study would seek to optimize sphere material, and geometrical properties such as radius *a* and spherical thickness, vis-a-vis manufacturing costs and biological constraints. The methods described in Kargl and Marston (1990) would be used for this purpose. The goal of this study would be to produce precise specifications of the prototype sphere, and predictions of *TS* enhancement. Also the issue of damping due to the sphere being encapsulated in fish tissue would need to be addressed; in the present feasibility study we have assumed this to be a small effect.

The experimental test would confirm the results of the parameter study. The test could be made at the Applied Physics Laboratory's Research Barge (R/V Henderson). A simple cage of the type described in Weibe, et al. (1990), which was used to measure the *TS* of encaged zooplankton, could be used for this purpose. The *TS* of both individual fish, and groups of fish, with and without implanted spheres would be measured in order to demonstrate final proof-of-concept.

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# A STUDY TO DETERMINE THE BIOLOGICAL FEASIBILITY OF A NEW FISH-TAGGING SYSTEM, PART II:

## Development of an Extended-range PIT-tag Interrogation System for Adult Salmon

## PROGRESS REPORT 1994-1996

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## **EXECUTIVE SUMMARY**

This report covers work performed by the National Marine Fisheries Service (NMFS) between 1994 and 1996 to develop a passive integrated transponder tag (PIT-tag) system to interrogate volitionally migrating adult salmon. A phased approach was adopted to realize this goal. The plan was to address basic electronic, mechanical, and hydraulic engineering questions in Phase I. In addition, several biological studies were initiated to provide basic information needed to design interrogation systems that minimize the potential risks to migrating adult salmon. The results obtained in Phase I will provide direction for the Phase II work involving the actual design and development of the extended-range PIT-tag interrogation system. Finally, Phase III will evaluate the biological responses of fish to the final design(s).

Phase I of the project encompassed six work elements, four of which our are covered in this report. These four elements were:

- 1) Evaluation of alternative electronic equipment to improve operational performance of the existing 400-kHz PIT-tag system.
- 2) Evaluation of 125- to 135-kHz PIT tags to determine if they had improved sufficiently to satisfy the longer read distance needed for an extended-range interrogation system.
- 3) Evaluation of how fish-ladder interrogation units could be designed to satisfy engineering and biological concerns.
- 4) Evaluation of fish behavior at three potential interrogation locations in a fish ladder using video cameras.

NMFS contracted RF Engineering to evaluate alternative electronic equipment to improve the operational performance of the existing 400-kHz PIT-tag system. They recommended using Helmholtz coils and more efficient C-Class signal amplifiers. NMFS concluded that even with the suggested modifications, the overall limitations of the 400-kHz system, especially its allowable radio-frequency (RF) emissions level, make it less likely to work for interrogating adult salmon than interrogating systems operating at lower frequencies (125- to 135 kHz).

A requirements document was prepared that described basic attributes and performance criteria of PIT tags needed for monitoring adult salmon within the Columbia River Basin (CRB). The evaluation of 125-to 135-kHz tags yielded some that met the performance criteria.

Simultaneous to NMFS' tag evaluation, a technical committee was formed within the International Standards Organization (ISO) to develop ISO standards for small implantable transponders and related transceivers. One of the tags that had satisfied the CRB performance criteria also fundamentally met the proposed ISO standard according to the manufacturer. This factor, in addition to the inherent advantages that an established standard normally brings such as product compatibility, competitive pricing, and multiple product sources led NMFS and others to recommend that the ISO standard be considered for the CRB. NMFS also recommended that the initial focus of the ISO-based system for the CRB should be on the development of an interrogation system for juvenile salmon. The knowledge gained from this development could then be applied to development of an adult system.

NMFS contracted the engineering firm Summit Technology to design several types of PIT-tag interrogation-coil housings that could be installed in select areas of fish ladders and specifically in the Fisheries Research Engineering Laboratory fish ladder at Bonneville Dam. This laboratory could then be used in the future to test the coil housings and the other components of an extended-range system. The engineers had to take biological, structural, hydraulic, and operation and maintenance concerns into account in their designs.

NMFS contracted the U.S. Army Corps of Engineers (COE) to use video cameras to monitor fish behavior in the Washington fish ladder at Bonneville Dam. Video equipment was deployed in an underwater orifice, in an overflow weir, and in a vertical slot. An informational baseline was created on behavior, swimming velocity, and orientation of fish passing through these three potential interrogation points. This information will permit a before and after comparison if PIT-tag interrogation equipment is installed in the ladder.

Data analysis of fish velocity showed that mean upstream velocity varied with species, within species, and by location. The mean upstream velocity ranged from 0.56 to 2.67 m/sec. Most fish swam directly through the three potential interrogation points and thus exposure to electromagnetic fields will likely be of short duration (seconds to minutes) for migrating adult salmon. The baseline information obtained will also aid in the design and development of extended-range PIT-tag interrogation systems to monitor adult salmon in CRB fish ladders.

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## INTRODUCTION

In the late 1980s, fisheries agencies recommended that the Bonneville Power Administration (BPA) install passive integrated transponder-tag (PIT-tag) interrogation systems at dams to interrogate migrating adult salmon. Restricted fish passage points (i.e., submerged orifices, overflow weirs, and vertical slots) within fish ladders were identified as possible PIT-tag interrogation points. In 1990, National Marine Fisheries Service (NMFS), under contract to BPA, initiated a project to investigate the technical and biological requirements of these larger interrogation systems. The initial tests were conducted using 400-kHz transceivers with 91-cm by 61-cm rectangular coils (tag excitation/receive antennas)(Prentice et al. 1994). We believed a PIT-tag interrogation system of this size could monitor most areas of interest. However, results showed that radio frequency (RF) emissions from the equipment exceeded acceptable limits established by the Federal Communications Commission (FCC) and ambient electrical noise significantly interfered with tag detection.

Based on current technology, we concluded that the practical reading range of the 400-kHz interrogation system was approximately 30 cm for a pass-through (tunnel-type interrogation system). We further concluded that a 30-cm system was too restrictive for use with adult salmon passing volitionally through PIT-tag interrogation systems.

During 1992 and 1993, NMFS sought proposals from PIT-tag manufacturers to develop an extended-range interrogation system that could be installed in adult fish-ladders located within the Columbia River Basin (CRB). The systems proposed were all based on 125- to 135-kHz operating frequencies. Unfortunately, based on technical or cost considerations, none of the proposals was satisfactory. However, this time period did mark the beginning of some significant improvements to the 125- to 135-kHz PIT tags as manufacturers started to incorporate new technologies (e.g., improved computer chips and tag coils). Therefore, after consultation with BPA in 1994, NMFS started a phased approach toward developing an extended-range PIT-tag system using in-house and outside resources.

The plan was to address basic electronic, mechanical, and hydraulic engineering questions in Phase I. In addition, several biological studies were initiated to provide basic information needed to design interrogation systems that minimize potential risks to migrating adult salmon. For example long exposures to electromagnetic fields might be a risk and thus, one study examined how long fish were exposed in fish ladders and another whether long exposures were harmful. The results obtained in Phase I will provide direction for the Phase II work that will address the actual design and development of the extended range PIT-tag interrogation system. Phase III will evaluate the biological responses of fish to the final design(s).

Six work elements of Phase I were completed during 1994-1996:

- 1) Evaluation of alternative electronic equipment to improve operational performance of the existing 400-kHz PIT-tag system.
- 2) Evaluation of 125- to 135-kHz PIT tags to determine if they had improved sufficiently to satisfy the longer read distance needed for an extended-range interrogation system.
- 3) Evaluation of how fish-ladder interrogation units could be designed to satisfy engineering and biological concerns.
- 4) Evaluation of fish behavior at three potential interrogation locations in a fish ladder using video cameras.
- 5) Evaluation of potential effects of electromagnetic fields (EMF) on maturing fish.
- 6) Evaluation of different approaches to improve tag retention in maturing salmon.

A discussion of work elements 1-4 is presented in this report, while work elements 5 and 6 will be included in a separate report that covers the biological studies completed during 1994-1996.

## EVALUATION OF ALTERNATIVE ELECTRONIC EQUIPMENT TO IMPROVE THE PERFORMANCE OF THE 400-kHz SYSTEM

In 1994, NMFS contracted RF Engineering to determine if alternative exciter and receive antenna designs were available that could overcome several performance limitations (e.g., limited read range, sensitivity to water-level changes by the antenna system) of the 400-kHz PIT-tag system used in the CRB. At the time of the contract, NMFS was reviewing whether the performance of the 400-kHz system could be improved sufficiently to meet the need for an interrogation system for volitionally migrating adult salmon.

## Contract activities included:

- 1) measuring the excitation sensitivity of Destron-Fearing PIT tags.
- 2) measuring the H-field (the part of electromagnetic field that energizes the tag) of a standard 400-kHz antenna.
- 3) designing a Helmholtz coil (antenna) for a 30.5-cm-diameter pipe.
- 4) constructing and evaluating ferrite-based excite and receive antennas.
- 5) measuring the effects of PIT-tag orientation on reading range.

A 30.5-cm diameter pipe was used simply as a point of reference, as larger-diameter antennas are required for interrogation of volitionally migrating adult salmon. A full description of these activities is presented in Appendix A.

Below is a summary of the findings by RF Engineering:

- 1) The minimum excitation level for 400-kHz tags was approximately two times greater in air than in water.
- 2) Under the test conditions employed by RF Engineering, the standard 400-kHz antenna system produced a lower H-field than expected.
- With both coils unshielded, a Helmholtz coil produced a near-field H-field which was double that produced by the standard 400-kHz antenna with similar far-field emissions. The Helmholtz coil also showed resistance to detuning from water agitation and nearby metal objects.
- 4) It was suggested that further improvements in system performance could be obtained by using more efficient C-Class signal amplifiers with the PIT-tag interrogation system.

- 5) Shielding the Helmholtz antenna significantly decreased its E-field emissions compared to standard 400-kHz antennas. Therefore, RF Engineering suggested several shield designs to further improve shielding efficiency.
- Tag orientation caused fewer detection problems when using a Helmholtz antenna.
- 7) The addition of ferrites to the exciter antenna decreased H-field attenuation.
- 8) It was suggested that gains in tag reading performance could be expected by separating the excitation and receive antennas.

These preliminary results suggest that it would be possible to make some improvement to the operating performance of the 400-kHz PIT-tag system. However, the extent of a possible improvement is unknown at this time without further laboratory and field tests because most of RF Engineering's tests were conducted under conditions quite dissimilar to those in the field or at the dams. Furthermore, NMFS believes that even with the suggested modifications implemented, the overall limitations of the 400-kHz system, especially its allowable RF emissions level, make it less likely to successfully detect adult salmon than interrogation systems operating at lower frequencies (125- to 135 kHz). Finally, several of the suggestions offered (e.g., C-Class signal amplifier and Helmholtz antenna) in this report could potentially be applied to these other systems.

## **EVALUATION OF 125-135 kHz PIT TAGS**

In January 1994, a requirements document was prepared by NMFS and other fisheries agencies that described basic attributes and performance criteria for PIT tags needed for monitoring adult salmon within the CRB. This document and a request for technical information was sent to all known manufacturers of 125-135-kHz PIT tags. These lower frequency tags require less energy to activate than 400-kHz tags and consequently have longer read ranges. The resulting technical information was evaluated by NMFS. Those manufacturers having PIT-tag technology that appeared to meet the performance criteria were asked to participate in a product evaluation. The PIT tags were evaluated on the basis of physical dimensions, read distance, read speed, code error rate, and level of electromagnetic energy required for activation. Initial product evaluations by NMFS personnel and an outside contractor were completed in June 1994. Details of the tag analyses are not presented here because they contain proprietary information. However, several of the tags satisfied the performance criteria.

During the period the tags were being evaluated, a committee from the International Standard Organization (ISO) was formed to develop standards for small implantable transponders (PIT tags) and related transceivers (tag energizing and receive equipment). An ISO standard (described in published documents ISO 11784 and ISO 11785) was ultimately approved by the committee in February 1995 and signed in 1996.

One of the tags that satisfied the performance criteria fundamentally met the proposed ISO standard according to the manufacturer. This factor, in addition to the advantages that a standard normally brings to a product (e.g., product compatibility, competitive pricing, multiple product sources) led NMFS and others to recommend that the ISO standard be considered for the CRB. In addition, several manufacturers indicated in 1994 that ISO-based portable transceivers and 12-mm tags suitable for fisheries applications would be available prior to or soon after the signing of the ISO standard.

Because of the interest and support shown by the international PIT-tag user community toward the formation of an ISO standard as well as the advantages that an ISO-based system could offer over the existing 400-kHz system used in the CRB, NMFS recommended to BPA that long-term efforts be directed toward the development of an extended-range PIT-tag system using a ISO-based system. This recommendation was made even though some non-ISO systems could also have been adapted for fisheries applications. Furthermore, it was jointly concluded by NMFS and BPA that if the ISO-based system were to be incorporated into the CRB, that efforts should first be focused on the evaluation and implementation of a juvenile system before undertaking the development of an adult system. We believed that much of the knowledge gained in development of a juvenile system would be applicable to the development of an adult system and that this course of action would result in reduced system development time and cost. In addition, this approach would provide time for manufacturers to develop new products that could perform under the more demanding conditions needed for interrogation of migrating adult salmon.

# ENGINEERING DESIGNS FOR INSTALLING PIT-TAG INTERROGATION EQUIPMENT IN FISH LADDERS

The design of a PIT-tag interrogation unit for use in adult fish ladders requires that a number of factors be considered: hydraulics, structural integrity of the ladder, ladder maintenance, fish passage, expected PIT-tag interrogation equipment performance, equipment accessibility, and overall system operation and maintenance. In addition, any design would need to be evaluated before it could be installed into CRB fish ladders. A contract was issued in 1995 to a structural engineering firm, Summit Technology, to:

- 1) Design several extended-range PIT-tag interrogation coil housings that could be used in fish ladders.
- 2) Examine a facility for the purpose of installing and evaluating such antenna housings.

Summit Technology personnel visited Bonneville Dam and then designed coil housings that could be installed and evaluated in the Fisheries Engineering Research Laboratory (FERL), otherwise known as the Washington Shore Experimental Fish Ladder. The firm's report describing the design of the coil housings and necessary modifications to the FERL fish ladder is presented in Appendix B.

Below is a summary of the findings by Summit Technology:

- 1) Coil housings could be manufactured that take into account the biological, structural, hydraulic, and operation and maintenance concerns.
- 2) Necessary modifications to the FERL could be made without jeopardizing the ladder's integrity.
- 3) Coil housings similar in design to those described could be deployed in the vertical slots, underwater orifices, and overfall weirs at other CRB fish ladders.

## VIDEO DOCUMENTATION OF FISH BEHAVIOR IN FISH LADDERS

Any PIT-tag interrogation system installed in fish ladders must be designed to minimize its impact on fish passage and ladder operation. To obtain needed design information, NMFS contracted the Portland District of the U.S. Army Corps of Engineers (COE) to use video cameras to monitor fish behavior in the Washington Shore Fish Ladder at Bonneville Dam. Video equipment was deployed in an underwater orifice, an overflow weir, and a vertical slot. The contract's objectives were as follows:

- 1) Document and establish fish behavior in a fish ladder before the installation of PIT-tag interrogation equipment.
- 2) Document orientation of fish (i.e., tag orientation) while passing through the three potential interrogation points;
- 3) Determine fish passage time (velocity) past the potential interrogation points; and
- 4) Estimate the potential time fish could be exposed to the electromagnetic field generated by the PIT-tag system.

The behavior of 728 salmon and other species (e.g., lamprey, shad) was recorded. Pacific lamprey (*Lampetra tridentata*) were of interest because their attaching behavior could potentially result in long exposure to the interrogation system's electromagnetic field. Furthermore, if lamprey were PIT tagged and they attached to the interrogation system, this would preclude other PIT-tagged fish from being read because two tags cannot be read simultaneously in the same electromagnetic field. The COE report for this study is presented in Appendix C. Below is a summary of the findings by the COE:

- 1) The video documentation successfully established a baseline that could be used to compare fish behavior before and after any fish ladder modifications.
- 2) Review of the video tapes showed that fish or tag orientation was satisfactory for PIT-tag interrogation at the three locations (underwater orifice, overflow weir, and vertical slot).
- 3) Results also showed that mean fish velocity varied greatly by species, for fish within species, and by location. Mean upstream fish velocities ranged from 56 cm to 267 cm/sec.
- Finally, the data showed that exposure time to the electromagnetic field would be short for most fish (seconds to minutes). However, lamprey were seen attached to the sides of underwater, orifice overflow, and vertical slots. This potential problem would need to be addressed in the design of PIT-tag interrogation systems deployed in fish ladders where PIT-tagged lamprey were present.

## SUMMARY, CONCLUSIONS, AND RECOMMENDATIONS

- 1) NMFS decided on a phased approach to develop an extended-range PIT-tag interrogation system using in-house and outside resources. In Phase I, basic electronic, mechanical, and hydraulic engineering questions were addressed. In addition, several biological studies were initiated to provide basic information for the design of interrogation systems that minimize potential risks to migrating adult salmon. Results from Phase I will provide direction for the Phase II work involving the actual design and development of an extended-range PIT-tag interrogation system. Phase III will then evaluate the biological responses of fish to this system.
- The evaluation of alternative electronic equipment for the 400-kHz system resulted in several recommendations to improve operational performance (e.g., Helmholtz coil designs, C-class signal amplifiers). However, NMFS believes that even with the suggested modifications implemented, systems operating at lower frequencies (125-135 kHz) would be more likely to detect adult salmon. Some of the suggestions offered in the review could potentially be applied to other PIT-tag interrogation systems.
- A requirements document was prepared that described basic attributes and performance criteria for PIT tags needed for monitoring adult salmon within the CRB. Evaluations of 125- to 135-kHz tags yielded some that met these performance criteria.
- 4) Simultaneous to NMFS' tag evaluation, a technical committee was formed to develop standards for small implantable transponders (PIT tags) and related transceivers. It appears the established standard will yield a system that will, in part, work for fisheries applications. Because of the advantages of a standardized product, NMFS recommended that the FDX-B ISO tag be considered for use in the CRB.
- NMFS also recommended that the initial focus of the ISO-based system for the CRB should be on the development of an interrogation system for juvenile salmon. The knowledge gained from this effort could then be applied to development of an adult system.
- NMFS issued a contract to the structural engineering firm Summit Technology to design PIT-tag coil housings that could be deployed and then evaluated in the FERL fish ladder at Bonneville Dam. The designed coil housings were for the underwater orifices, overfall weirs, and vertical slots of a fish ladder. The designs took into account biological, structural, hydraulic, and operation and maintenance concerns. Summit Technology's report indicated that the necessary modifications to the FERL could be made without jeopardizing ladder integrity. In addition, similar coil housings could be deployed in other CRB fish ladders.

- 7) NMFS contracted COE to use underwater video equipment to record fish behavior within the Washington shore fish ladder at Bonneville Dam. Video equipment was deployed in an underwater orifice, an overflow weir, and a vertical slot.
- Fish velocity and orientation while passing through areas of interest were video recorded. Data analysis showed that mean swimming velocity varied with species, within species, and by location. Mean fish velocity ranged from 0.56 m to 2.67 m/sec. Neither fish orientation nor distance from potential antenna locations (as they passed through the various structures of interest) appeared to be outside the limits of possible PIT-tag detection. Further, the data indicated that exposure time to the PIT-tag interrogation system's electromagnetic field would be minimal for most species (lamprey being the potential exception).

Baseline fish behavior information obtained using video will be used in the design of PIT-tag interrogation systems for adult fish and for evaluating potential biological impact of systems deployed in the fish ladder.

## **ACKNOWLEDGMENTS**

Support for this research came from the region's electrical rate payers through the Bonneville Power Administration.

## APPENDIX A

Final Report: 400 kHz PIT Tag Antenna System

RF Engineering

## FINAL REPORT

# 400 kHz PIT TAG Antenna System March 15, 1995

WORK PERFORMED FOR:

NATIONAL MARINE FISHERIES SERVICE
MANCHESTER, WA 98353

REF: 43ABNF402429 I

## PREPARED BY:

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## WORK DESCRIPTION (per PURCHASE ORDER)

Evaluate RF Engineering proprietary antenna and receive system in reference to the Destron/Fearing 400 kHz Pit Tag System used in the Columbia. River basin.

This Study was funded in two parts:

- 1. Initial Study: Statement of Work is attached as Appendix E
- 2. Extension: Statement of Work is attached as Appendix E1

## **OVERVIEW**

This study was undertaken to determine if alternative exciter and receiver antenna designs were available which could improve the performance of the existing 400 kHz Pit tag antenna systems.

The present 400 kHz Pit tag tunnel (2U:3 by 61 cm) has several limitations:

- 1. Unit is difficult to tune and detunes easily due to:
  - 1. Amount of water in tunnel
  - 2. Water turbulence
  - 3. Metal in surrounding structure (i.e. concrete re-bar)
- 2. Maximum read range is 20.3 cm (8") and is heavily dependent on Pit tag orientation.

(Read range limitation appears to be due to the receive sensitivity rather than the excite power)

- 3. Receiver is sensitive to external interference.
- 4. Tunnel must be made of non-conducting material.
- 5. Exciter loop must enclose the tunnel and cannot be used in a passby mode.
- 6. The driver circuitry requires high voltages and high current.
- 7. Exciter has high emission levels and is difficult to shield due to detuning effect.

An ideal Pit tag system would resolve the limitations discussed in the prior paragraph and would also satisfy the requirements listed below. These additional issues are not addressed in this study. They are presented here in order to provide a more complete list of the fisheries service requirements.

- 1. Able to read in both juvenile and adult measurement environments.
- 2. Able to read multiple fish (either by increasing range or read speed)
- 3. Able to read in less than  $40~\mathrm{ms}$  and a minimum speed of  $20~\mathrm{fps}$ .
- 4. Able to detect and read fish heart beat.
- 5. Able to detect presence and size of non tagged fish.
- 6. Meets regulatory and safety requirements FCC emissions ANSI Safety Biologically Safe for animals (considerations are: Frequency, CW vs Pulse, Field Strength, etc)
- 7. Readers (including portable units) would be compatible with all Pit Tags, including future ISO Standards.

This study was undertaken to determine if new antenna/exciter designs would overcome same of the limitations of the present 400 kHz Pit tag tunnel. Two different antenna systems were studied and a review of other potential antennas was performed. These different systems and their primary benefits are listed below:

- 1. Ferrite based Exciter and Receive Antennas.
  - 1. Good passby candidate
  - 2. Can develop orthogonal H fields
  - 3. Can be used in conductive channels.
- 2. Helmholtz Coil Exciter and Ferrite Receive Antenna.
  - 1. Very uniform H field.
  - 2. Active length can be made any length (increases minimum read velocity).
  - 3. Insensitive to detuning
  - 4. Low exciter voltage.

3. Reviewed additional antenna types for possible exciter use.

(Review only - no analysis or measurements were performed)

- 1. Compact Loop antenna with integral ground plane.
- 2. Microstrip type launchers (with ground plane).
- 3. Solenoid structures.

  Generally sate performance and magnetic field geometry as Helmholtz coil.

NOTE: Items 3.1 and 3.2 have potential for use in passby applications and for orthogonal H field excitation.

## ACTIVITIES

## 1. MEASURED EXCITATION SENSITIVITY OF DESTRON/FEARING PIT TAG

#### DISCUSSION

Measurements were made of the minimum H field required to excite a Destron/Fearing Pit Tag over the frequency range 200 to 800 kHz. Results are for a single tag and are measured in water and air.

RESULTS are given in Appendix B.

#### COMMENTS

Selected results for a tag in water are:

- 13 Amp-T/meter at 400 kHz
- 8 Amp-T/meter at 600 and 800 kHz.

Minimum excitation levels in air are approximately two times greater than in water.

## 2. MEASURED H FIELD OF THE NMFS PROVIDED 400 KHZ TUNNEL

RESULTS are given in Appendix A.

- 1. Nominal H field: 30 Amp-Turns/meter.
- 2. H field ranges from 18 to 20 Amps-T/meter over the central plane of the antenna aperture.

#### COMMENTS

The above H field results are considered to be low compared to a properly tuned exciter coil and driver (per NMFS staff). Discussions during the October 27, 1994 meeting at RF Engineering suggested that desired excitation levels were 40 Amp-T/meter minimum. Whit Patton suggested that 80 Amp-T/meter would be a better value.

Proposed 1991 ANSI H field (and E field) safety guidelines are given in Appendix F1. They suggest an acceptable H field limit for human exposure at 400 kHz to be 40 Amp-T/meter.

## 3. DESIGNED HELMHOLTZ COIL EXCITER (for 12" PVC Pipe)

## DISCUSSION

Magnetic coils (such as the Helmholtz and solenoid coils) provide a uniform H field over the entire interior region of the coil. This is in contrast with small antennas (such as ferrite coils) whose H field falls off inversely with the cube of range.

The physical operation at a magnetic coil requires that the magnetic flux flow inside and <u>outside</u> the coil. The need for this external flux path suggests that these coils are best used with <u>non-conductive</u> pipes and tunnels.

External metal shields can be used to reduce radiated emissions; however, they must be carefully designed in order to avoid detuning the coil.

## ANTENNA/DRTVER DESCRIPTION

Antenna (Helmholtz Coil):

Two coils (2 turns each) spaced 6" apart.

Mounted an outside of 12" plastic bucket (simulated PVC pipe)

## Antenna matching network:

Coils are connected in parallel and are driven from a capacitive step up network.

Antenna circuit is tuned to 400 kHz.

Coil voltages range from 60 to 200 volts peak to peak.

## Exciter Driver #1:

Low impedance Class B Power Amplifier operating at 400 kHz.

Power supply:  $\pm$  20 Volts at 200 mA.

Amplifier schematic is given in Appendix C1.

## Exciter Driver #2:

Goal was to provide a times 2 increase in H field compared to Driver #1 and to determine if a balanced differential antenna drive would result in lower radiated emissions.

## Design consisted of:

- 1. Redesigning the antenna matching network for a balanced input.
- 2. Use an input transformer and two Class B amplifiers to provide the required balanced output voltage.
- 3. Amplifier schematic is given in Appendix C3.

#### RESULTS:

Exciter #1: Measured H field measurements are given in Appendix C. H field ranges from 14 to 20; Amp-T/meter at the central plane of the antenna.

(See discussion on desired H field on page 6, section 2)

Exciter #2: See Appendix C2. H field has approximately doubled.

#### COMMENTS

Exciter Driver #2 was designed as a balanced amplifier in order to decrease far field emissions and to double the output drive of Exciter Driver #1. Emission measurements did not show any improvement due to this balanced drive (see section 4) and increased drive capability could be more effectively achieved by paralleling the output of the drivers using a power tranformer. Paralleling the driver outputs provides a lower source impedance and simpler antenna matching network.

Exciter #1 and #2 were designed as class B amplifier stages in order to provide:

- 1. Good power amplifier efficiency
- 2. Low harmonic distortion

(in order to reduce radiated harmonics).

Emissions testing indicated very low harmonic radiation from the antenna matching networks and their associated Helmholtz coils. It is probable that Class C amplifiers may be used with this system and that amplifier operating efficiencies up to 50% may be achieved. This would allow several benefits such as lower drive voltages or larger coil diameters (up to 48").

The Helmholtz coil circuits showed good resistance to detuning due to water agitation or nearby metal objects.

#### 4. MEASURED RADIATED E FIELD EMISSIONS

#### DISCUSSION

Measurements were made of the radiated emissions from the 400 kHz Pit tag tunnel and the Helmholtz coil at ranges of 30 and 100 feet using both E field and H field probes. The E field probes were 17" ground mounted monopoles, the H field probes were 8.25" diameter loops.

The E field measurements were erratic in signal level both inside and outside the building, results were location sensitive and did not behave well with range. H field measurements were in general far more consistent both inside and outside and behaved well with range. In addition H field measurements and calculations were in good agreement for the 12" Helmholtz coil (see Appendix G).

H field measurements were converted to E field equivalents before graphing in order to compare them with FCC emissions limits.

NOTE: Measuring H field and then calculating E field from the measured H field value is a common practice in this frequency range and is an approved FCC technique.

#### RESULTS

Radiated E field emissions for the Pit Tag tunnel and the 12" Helmholtz coil with Driver #1 are given in Appendix G. Emissions are shown for the following Exciter configurations:

- 2. RFE System w/NO Shield unshielded Helmholtz coil. (H field inside coil: 14 to 20 Amp-T/meter)
- 3. RFE System w/Open Shield shielded Helmholtz coil. (Copper shield, 1 open turn, 14" wide, 8" separation from coil. Same H field as item 2 above)
- 4. RFE System w/Shorted Shield shielded Helmholtz coil. (Copper shield, 1 shorted turn, 14" wide, 8" separation from coil. Same H field as item 2 above)
- 5. RFE System Calculated calculated E field for unshielded Helmholtz coil.

No E field harmonics of the  $400\ \mathrm{kHz}$  were observed from the RFE Exciters.

#### COMMENTS

The 400 kHz Pit tag tunnel and the unshielded Helmholtz coil generated similar internal H fields, but E field emissions from the tunnel were approximately 2 times greater than for the Helmholtz coil. This difference could be due to the larger size of the tunnel (197 sq in) compared to the coil (113 sq in) and is not deemed significant.

The calculated and measured values of E field emissions for the Helmholtz coil were consistent at the measured ranges of 30 and 100 feet. Extending this correlation to 300 meters allows a projection of E field emissions at the FCC specified measurement range for both the Pit tag tunnel and the various Helmholtz coil configurations.

The projected values of E field emissions at 300 meters are shown in Appendix G. The Pit Tag tunnel E field is projected to be 38 uV/meter versus an FCC limit of 6 uV/meter (see Appendix F). The various Helmholtz coil configurations give E field values ranging from 12 uV/meter to .23 uV/meter thus showing the benefits associated with shielding the coil.

## HELMHOLTZ COIL SHIELDS

It is expected that an electrostatic shield or a shorted copper shield will be used to reduce far field emissions to FCC specified levels.

The shorted turn provides magnetic and electric shielding and reduces interactions with ferrous structural materials. It may require more space than an electrostatic shield.

#### 5. BUILT FERRITE BASED EXCITER ANTENNAS

#### DISCUSSION

Ferrite based exciter antennas are expected to have the following benefits:

- 1. Exciter antennas can be placed at right angles to each other. This reduces the sensitivity to Pit tag orientation.
- 2. Ferrite antenna are not sensitive to detuning or nearby non-ferrous conductive materials. They can be located inside a non-ferrous conductive tunnel.
- 3. May be used in passby mode.
- 4. Require low excitation voltage.

The primary concern with this type of exciter structure is that unlike the Helmholtz Coil or solenoid structures it does not produce a uniform H field. The H field of the ferrite antennas falls off as range cubed and to achieve acceptable H field levels in the center of the 400 kHz Pit tag tunnel requires very high H field levels at the tunnel walls.

#### RESULTS

#### Experiment #1

Measures the H field in a large tank of water far the following conditions: (see Appendix D1)

- 1. Single ferrite, 2.5" long
- 2. Single ferrite, 7.5" long
- 3. Two ferrites, spaced 8" apart
- 4. Single ferrite inside a conductive ring.

Results are given in Appendix D

#### Experiment #2

Measurements in the H field in a large tank of water for the following conditions: (see Appendix D3)

- 1. Single ferrite, 2.5" long
- 2. Two ferrites, one above the other
- 3. Two ferrites, spaced 8" apart
- 4. Four ferrites, two by two.

Results are given in Appendix D2

#### COMMENTS

A review of the data in Appendix D and D2 shows that the H field for a single ferrite exciter decreases roughly by a factor of 100:1 over a range of 0" to 4". This can be decreased to a factor of 40:1 when two ferrites are spaced 8" apart. The effect of the conductive Brass ring is seen to increase the overall H field by a factor of 2, but does not change the attenuation versus range profile.

It is expected that 4 ferrites in an 8" diameter round pipe would improve the attenuation ratio to 20:1. This would give an H field profile ranging from 20 Amp-T/meter at the center of the pipe to 400 Amp-T/meter at the pipe edge. This might be an effective arrangement inside non-ferrous metal pipe, particularly if a second set of ferrites were placed at right angles to the original set in order to reduce the sensitivity to Pit tag orientation.

#### 6. BUILT FERRITE BASED RECEIVE ANTENNAS

#### DISCUSSION

The Destron/Fearing Pit tag is excited at 400 kHz and responds at 40/50 kHz. It is expected that separating the transmit and receive functions will allow optimization of each antenna and in particular it should allow an improvement in the read range.

#### HARDWARE DESCRIPTION

Exciter - operates at 400 kHz

Receiver - operates at 40/50 kHz
Antenna: Single 40 turn Ferrite antenna.

(Two may be required in final system)

Amplifier: Low noise amplifier with two 45 kHz bandpass filters (40 to 50 kHz) and a 400 kHz notch filter. (Schematic is shown in Appendix H1)

#### RESULTS

Measurement were performed by mounting a Pit tag on a small ferrite exciter: the exciter signal level was set to a level just large enough to cause the Pit tag to respond with the normal FSK response signal (40 to 50 kHz).

The receive antenna was connected to the amplifier discussed above and measurements were made of relative signal response at the output of the amplifier. Results are shown in Appendix H for relative response in water and air versus distance from the receive antenna. Results were not highly dependent on Pit tag or receive antenna orientation.

A second set of tests were run with the ferrite based receive antennas in the 12" PVC pipe using the Helmholtz coil exciter. Return signals from the Pit tag in this test could be read up to a range of 6" from the receive antenna. The use of two (or three) receiving antennas spaced around the pipe would provide an adequate receiving antenna system.

#### COMMENTS

The primary restriction on receiver range in the measurements discussed above was an interfering 45 kHz signal in the middle of the receiver passband. The source of the spurious 45 kHz signal was not identified, but it was a stable signal which could be measured throughout the lab area. It is likely that this spurious signal would limit the overall system dynamic range even after installation of the normal emissions shield.

The signal strength and H field orientation of the spurious 45 kHz signal was measured and was found to be very consistent over the measurement area. It is projected that a second set of receive antennas placed 12" to 18" away from the Pit tag could be used to cancel the interfering signal in the primary receiving antennas without reducing the desired Pit tag return signal.

#### 7. MEASURED READ DISTANCE VERSUS PIT TAG ORIENTATION

#### DISCUSSION

The Helmholtz coil exciter and ferrite based receive antennas discussed in Sections 3 and 6 were used to measure the effect of Pit tag orientation on system read range.

The measurements were performed at three different H field levels, at varying ranges and at different locations inside the 12" diameter pipe.

RESULTS are given in Appendix 1

#### COMMENTS

The Pit tag could be rotated 50 degrees from the optimum orientation and still be read throughout the Helmholtz coil area when the H field was 30 Amp-T/meter. The minimum read angle decreased to 40 and 35 degrees when the H field strength was reduced to 22 and 16 Amp-T/meter respectively. These minimum readings occurred towards center of the pipe.

The above results could be improved by the following means:

- 1. Increase the H field to 40 Amp-T/meter as discussed in Section 3 of this report
- 2. Adjust the geometry of the Helmholtz coil in order to generate orthogonal H fields inside the pipe.

The following are alterations which could be made to the Helmholtz coil in order to generate orthogonal H fields inside the 12" PVC pipe.

- 1. Increase the number of coils and place them at 30 or 45 degree angles relative to the pipe axis.
- 2. Place additional coils on the outside surface of the pipe, this will create an H field transverse to the major axis of the pipe.

#### RECOMMENDATIONS

The present 400 kHz Pit tag tunnel uses a single solenoid type coil which serves as both a tuned 400 kHz exciter coil and a tuned 40 to 50 kHz receive coil. The system has several limitations which are discussed on page 3 of this report.

This study was undertaken to determine if alternative antenna designs were available which could improve the performance of the present 400 kHz system, particularly with respect to the following applications:

- Circular Pipes (or Rectangular tunnels) (non conductive channel)
- 3. Passby Applications
   (both conductive and non-conductive channel)

# 1. Circular Pipes (or Rectangular tunnels) -non conductive channel

#### EXCITER

The Helmholtz coil described in this report has a number of benefits for an application with a non-conductive channel. Some of those benefits are listed below:

- 1. Uniform H field
- 2. Insensitive to detuning due to water level, shielding materials, etc.
- 3. Coil length can be increased for higher read velocity.
- 4. Coil geometry can be modified for orthogonal H fields.
- 5. Very large openings can be excited (up to 48" diameter) due to low coil impedance.
- 6. Simple excites amplifiers (low voltage and easy parallel operation).

#### RECEIVER

The ferrite based receive antennas discussed in section 6 can be integrated into very sensitive receiver systems. The ability to separate the excite and receive antennas allows this optimization to occur.

The receive system dynamic range can be increased by the use of a second set of interference sensing antennas:

#### 2. Rectangular Tunnels (maximum 8" wide) -conductive channel

#### EXCITER

The ferrite based exciter antennas discussed in section 5 can be used in 8" wide conductive channels since the tails can be operated on top of a conductive ground plane. H field in this application would vary 40:1 (i.e. 20 to 800 Amp-T/meter). This type of exciter is best used in an area with high stream flow in order to limit animal H field exposure.

Alternative antennas (i.e. loop antenna or microstrip antenna with integral ground planes, see page 5) can also be considered as excites antennas for this application.

#### RECEIVER

The ferrite based receive antennas discussed above can be used as receivers in a conductive channel since they can be operated with a ground plane.

# 3. Passby Application (Shelf, 6 ft wide by 12" high) -

#### EXCITER

The ferrite base excites antennas discussed in item 2 above can be used in passby applications; however, they have a maximum range of approximately 4" with an expected H field variation of 100:1. The alternative antennas discussed above and on page 5 (i.e. loop antenna with integral ground plane or microstrip antenna with ground planes) offer ranges greater than 4" with a likely range of 12" and an H field variation of 20:1.

The performance of these antenna types can be evaluated experimentally or by computer simulation (CAE). The primary challenge for this application is the 12" read height and not the 6 ft shelf width.

#### RECEIVER

Same as item 2 above.

#### POTPOURRI

The following is a list of items which have been discussed during this study. No actions or recommendations are suggested here, they are listed as a reminder only.

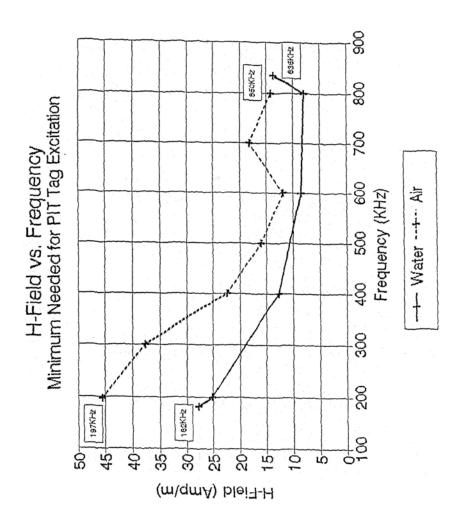
- 1 Operating the Destron/Fearing Pit Tag at 125 kHz.
- 2. Receiving the Pit Tag return signal as RF Sidebands (350 to 450 kHz frequency range) rather than as 40 and 50 kHz law frequency signals.
- 3. Plug and play packaging, software, etc.

Appendix A

H-Field Plot for NMFS 400 KHz Antenna System:

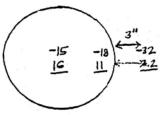
Top Top Top

Top	TOP	Top
-7 -10 -8	-8 -10 -8	-13 -12 -13
40 28 36	36 28 36	20 22 20
-12 -14 -10	-12 -14 -11	-14 -13 -14
27 18 28	22 18 25	18 20 18
7 -9 -7	-8 -10 -8	-14 -13 -13
40 32 40	35 20 36	13 20 20
Bottom	Bottom	Bottom
Plane of 10 Turn	Plane of 8 Turn	P, lane Between
Loop	Loop	Loops
Top  -35 -94 -38  1.6 1.8 1.1  -31 -30 -31  2.2 2.8 2.2  -35 -33 -36  1.6 20 1.4  Bottom	Top  -37 -38 -38  1.3 1.1 1.1  -25 -34 -33  1.6 1.8 20  -38 -36 -38  1.1 1.1 1.1  Bottom	Note: Top numbers are signal level in dBh measured with Spectrum Analyzer. Underlined numbers are derived H-field levels in Amp/m.
12" in Front	12" in Back	

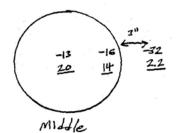


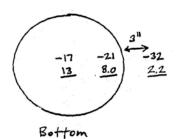
# Appendix C

H-Field Plot for RFE 400 KHz Antenna system:



Surface

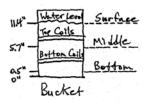




Note: Top numbers are signal level in dlem measured with Spectrum Analyzer.

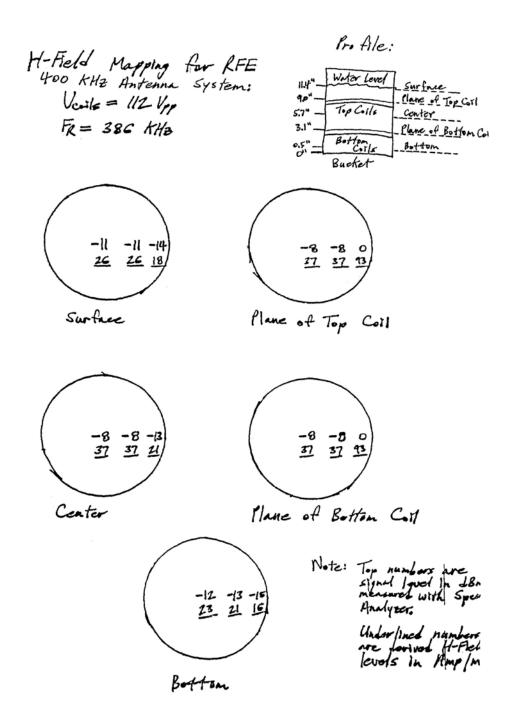
Underlined numbers are derived H-field levels in Amp/m.

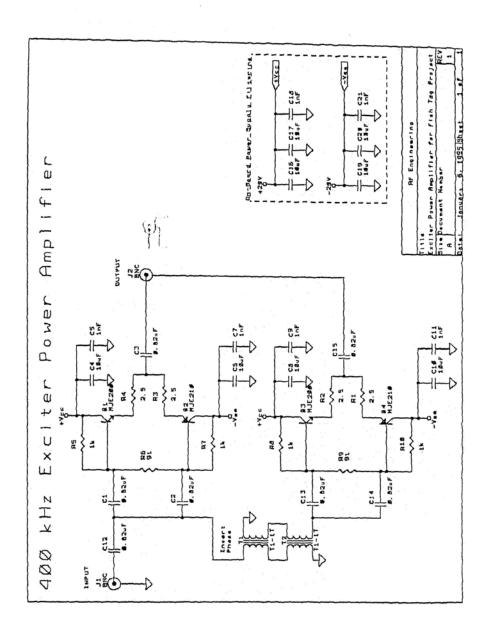
ProAle:



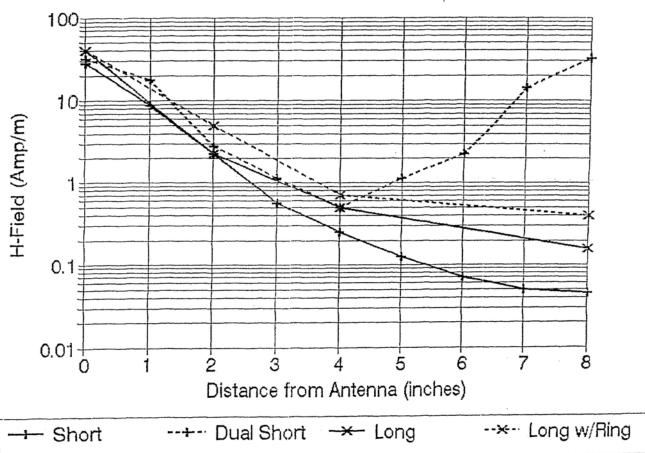
17 7 5 G PUSH-PULL UNITY GAIN POWER AMPLIFIER 2 22 200 292 0.0 8 등 ชรั้ -|-|> 20 200 92 TIP42 200 28 +280 83.57K 84 3.57× 383 . 82vF 8. 82uF 27 5 X INPUT

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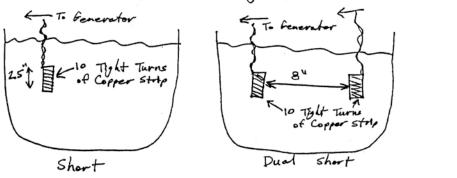


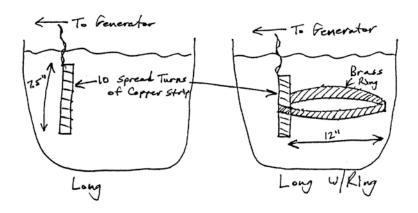


H-Field vs. Frequency Various Ferrite Antenna Configurations



Ferrite Antenna Configurations using Copper Strip Windings





Work Objectives and Specifications

## Objective:

Determine if RF Engineering's proprietary antenna and receive design will excite a Destron/Fearing 400 kHz passive integrated transponder (PIT) tag and receive the return signal.

Work Description and Expected Products:

1) The work will be conducted with Destron/Fearing 400 kHz PIT tags. Tag excitation and receive equipment will be a combination of RF Engineering and Destron/Fearing excite and read equipment where appropriate. NMFS will Furnish all Destron/Fearing tags, exciter, and receive equipment to RF Engineering for the tests. This equipment and excess tags will be returned to NMFS in its original configuration and working condition unless specified by NMFS.

#### 2) Read Distance:

- a) Using RF Engineering's proprietary antenna design and Destron/Fearing receive equipment, determine the maximum read distance at three power levels of RF Engineering's choice when the tag is in its optimal orientation, Record both the excitation and return signal strengths under this condition.
- b) Determine the maximum read distance when the tag is perpendicular, 45°, and parallel to the H field at one power level and provide an H field profile for each condition and estimated strength.
- 3) Determine the effect on both excitation and receive signal strength when grounded ferrous and non Ferrous metals are within 2.54 cm and 10 cm from the excitation and/or receive coil.
- 4) Destron\Fearing tags are reported to have a natural frequency around 900 kHz. Determine the upper frequency limit in which RF Engineering's proprietary antenna design can excite tags with performance greater than or equal to current technology.
- 5) Evaluate the present excitation and receive design of Destron/Fearing excitation and receive circuitry. Provide a report detailing how this design and present equipment can be integrated into the RF Engineering proprietary antenna and reader system to enhance overall read performance of the existing PTT tag system used in the Columbia River Basin.
- 6) A report will be prepared that includes a methods and materials and a results and discussion section. All test results will be presented in the report.

RF Engineering

Amendment: Work objectives and specifications

Work Description and Expected Products:

1) The work will be conducted with Destron/Fearing 400 kHz PIT tags. Tag excitation and receive equipment will be a combination of RF Engineering and Destron/Fearing excite and read equipment where appropriate. NMFS will furnish all Destron/Fearing tags, exciter, and receive equipment to RF Engineering for the tests. This equipment and excess tags will be returned to NMF5 in its original configuration and working condition unless specified by NMFS.

# 2). Increase power:

Increase the power level from, 20~V to 40~+~V to the Helmholtz coil and RF Engineering's proprietary antenna design. Determine H-field strength and maximum tag excitation/receive distance at the increased power levels.

# 3) Tag Read Range:

Determine the maximum excitation/receive distance underwater using multiple antenna (coil) configurations e.g., multiple excite coils with separate receive coils, multiple duel purpose coils, one large excite coil with multiple receive coils, etc.

#### 4) RF Emissions:

Determine RF emissions for the 400 kHz 20.3 cm by 61 cm Pit tag tunnel (unshielded) when energized with RF Engineering's excitation/receive system. The measurements will be For the fundamental and harmonic frequencies and conducted in accordance to FCC requirements. The measurements will be made at a minimum of 30 meters from the radiating source using a H-field loop antenna and appropriate spectrum analyzer:

# 5) Report:

A report will be prepared that includes a methods and materials and a results and discussion section. All test results will be presented in the report.

- § 15.209 Radiated emission limits; general requirements.
- (a) Except as provided elsewhere in this subpart, the emissions from an intentional radiator shall not exceed the field strength levels specified in the following table:

Frequency (MHz)	Field strength (microvolts/meter)	Measurement distance (meters)
0.009-0.490 0.490-1.705 1.705-30.0	2400/F(kHz) 24000/F(kHz) 30	300 30 30
30-88 88-216	100 ** 150 **	3
216-960 Above 960	200 ** 500	3 3

- \*\* Except as provided in paragraph (g), fundamental emissions from intentional radiators operating under this section shall not be located in the frequency bands 54-72 MHz, 76-88 MHz, 174-216 MHz or 470-806 MHz. However, operation within these frequency bands is permitted under other sections of this part, e.g., Secs. 15.231 and 15.241.
- (b) In the emission table above, the tighter limit applies at the band edges.
- (c) The level of any unwanted emissions from an intentional radiator operating under these general provisions shall not exceed the level of the fundamental emission. For intentional radiators which operate under the provisions of other sections within this part and which are required to reduce their unwanted emissions to the limits specified in this table, the limits in this table are based on the frequency of the unwanted emission and not the fundamental frequency. However, the level of any unwanted emissions shall not exceed the level of the fundamental frequency.
- (d) The emission limits shown in the above table are based on measurements employing a CISPR quasi-peak detector except for the frequency bands  $9-90\,\mathrm{kHz}$ ,  $110-490\,\mathrm{kHz}$  and above  $1000\,\mathrm{MHz}$ . Radiated emission limits in these three bands are based on measurements employing an average detector.
- (e) The provisions in Secs. 15.31, 15.33, and 15.35 for measuring emissions at distances other than the distances specified in the above table, determining the frequency range over which radiated emissions are to be measured, and limiting peak emissions apply to all devices operated under this part.
- (f) In accordance with Sec. 15.33(a), in some cases the emissions from an intentional radiator must be measured to beyond the tenth harmonic of the highest fundamental frequency designed to be emitted by the intentional radiator because of the incorporation of a digital device. If measurements above the tenth harmonic are so required, the radiated emissions above the tenth harmonic shall comply with the general radiated emission limits applicable to the incorporated digital device, as shown in Sec. 15.109 and as based on the frequency of the emission being measured, or, except for emissions contained in the restricted frequency bands shown in Sec. 15.205, the limit on spurious emissions specified for the intentional radiator, whichever is the higher limit. Emissions which must be measured above the tenth harmonic of the highest fundamental frequency designed to be emitted by the intentional radiator and which fall within the restricted bands shall comply with the general radiated emission limits in Sec. 15.109 that are applicable to the incorporated digital device.
- (g) Perimeter protection systems may operate in the 54-72 MHz and 76-88 MHz bands under the provisions of this section. The use of such perimeter protection systems is limited to industrial, business and commercial applications.

[54 FR 17714, Apr. 25, 1989; 54 FR 32339, Aug. 7, 1989; 55 FR 18340, May 2, 1990; 62 FR 58658, Oct. 30, 1997]

page

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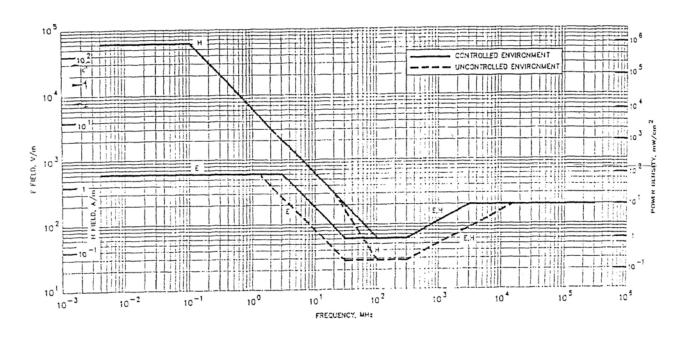
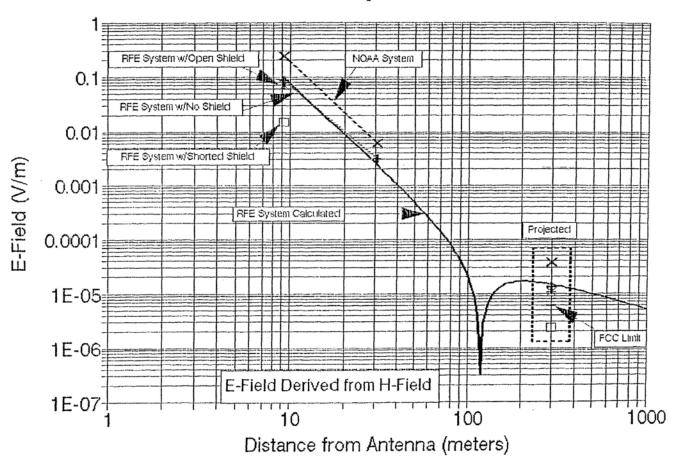


Fig. 4—Proposed 1991 ANSI RF protection guidelines for body exposure of humans.

FROM AARL Handbook

# E-Field Strength vs. Distance RFE and NOAA Systems at Fr=400KHz



APPENDIX

PIT Tag Return Signal Read Distance vs. Tilt Angle 1/11/95

F(Tx) = 400kHz F(Rx) = 45kHz

V(Tx): [Vpp] 112 81 58

h(Tx): [A/m] 29.9 21.6 15.5

Position:	Maximum	Tilt A	Angle	(degrees):	
Edge:	Top Coils	60		60	50
	Center	55		50	40
	Low Coils	60		60	50
4" Inside	Top Coils	60		60	45
	Center	50		45	35
	Low Coils	60		60	45
6" Inside	Top Coils	60		60	45
	Center	55		40	35
	Low Coils	60		60	45

# APPENDIX B

# Predesign Report: Prototype Testing of Passive Integrated Transponder (PIT) Tag Monitors for Adult Salmon

Summit Technology

# PREDESIGN REPORT

# PROTOTYPE TESTING OF PASSIVE INTEGRATED TRANSPONDER (PIT) TAG MONITORS FOR ADULT SALMONIDS

BONNEVILLE DAM
SECOND POWERHOUSE
NORTH SHORE FACILITIES

FEBRUARY 1996

# Prepared for:

- ☐ U.S. DEPARTMENT OF COMMERCE
- ☐ NATIONAL OCEANIC AND ATMOSPHERIC ADMINISTRATION
- ☐ NATIONAL MARINE FISHERIES SERVICE
- ☐ COASTAL ZONE & ESTUARINE STUDIES DIVISION

Prepared by:



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#### **EXECUTIVE SUMMARY**

National Marine Fisheries Service (NMFS), with support from Bonneville Power Administration (BPA), is investigating the technical feasibility of using Passive Integrated Transponder (PIT) tags to monitor the movement of adult salmonids passing through fish ladders. Although PIT tag transceivers are commonly used to track juvenile fish, this NMFS undertaking is unique in that it proposes to interrogate adult fish in fish ladders using a variety of PIT tag transceiver antenna sizes and geometries.

The program proposed by the NMFS includes several tasks. This design effort addresses two of those tasks.

Evaluate the technical feasibility of using Bonneville Dam's Fisheries
Engineering Laboratory fish ladder to study PIT tag transceivers and antenna
designs for the interrogation of adult fish.
Design PIT tag antenna housings that: a) enable a variety of antenna coil designs to be evaluated within a single housing, and b) can be deployed in an adult test facility without altering the facility's structural integrity, hydraulics, or modifying fish behavior.

Using engineering and hydraulic criteria, Bonneville Dam's Fisheries Engineering Laboratory (FEL) fish ladder was evaluated as to its suitability as a location to evaluate the proposed adult PIT-tag transceiver system.

Once the site is approved by all stakeholders, several modifications to the FEL fish ladder would be required to carry out the program. The ladder would be modified to emulate two fish passage areas, Vertical Slot and Orifice/Weir, of the Bonneville 2ND Powerhouse North Shore Fish Ladder. This ladder is considered typical of most ladder installations in the Columbia River Basin.

As part of this effort, antenna housings that emulate the Vertical Slot and Orifice/Weir area of a fish ladder are discussed. The antenna housings were designed to be installed and removed and not alter the ladder hydraulics or fish behavior. The design also enables various antenna configurations to be used within a housing for evaluation purposes.

The Vertical-Slot antenna housing was patterned after Baffles No. 15, 16, and 17 of the Bonneville 2ND Powerhouse, North Shore Fish Ladder, Exit Control Section. With similar opening width, opening height, and angle to flow, it is anticipated that the housing will not have fish passage and hydraulic performance different from the unmodified section of the existing North Shore Fish Ladder.

Upon completion of the adult PIT tag system evaluation by NMFS, anchor bolt sleeves that are installed in the floor and the sill will have plugs installed and left flush.



The Orifice/Weir antenna housings (based upon a combination of pass-through and pass-by concepts respectively) would be constructed as a full-scale model of Weirs 38 through 43 (of the Fisheries Engineering Laboratory Entrance Fish Ladder) and Weirs 44 through 48 (of the Fisheries Engineering Laboratory Exit Fish Ladder) (Fig. 2).

Conceptual designs have been prepared for two Orifice/Weir antenna housings, a Structural Fiberglass Orifice/Weir antenna housing constructed of fiberglass shapes and plates, and a Fiberglass Reinforced Concrete antenna housing. Of these two designs only one would be built.

A removable antenna housing would be installed as the top portion of the overflow weir; a second removable antenna housing, installed in a structural frame would serve as the orifice. Fish passage and hydraulic performance of the antenna housing should be similar to the existing Orifices/Weirs in the Fisheries Engineering Laboratory Entrance Fish Ladder and the North Shore Fish Ladder.

Upon completion of the test program, the Orifice/Weir antenna housings would be removed. The fiberglass anchor bolts would be cut off and reinforcing steel would be doweled into the walls and floor. Once the dowels are installed, the orifice/weir concrete would be replaced as shown in the original construction drawings, and any temporary bolt holes would be grouted flush.

The PIT-tag system operates using a strong electromagnetic field. If during the operation of either the Vertical-Slot or Orifice/Weir antenna system it is found the radio frequency (RF) emissions of the system exceed federal standards, an RF shield may be required. A conceptual design for this shield is provided.

Although this design report addresses the feasibility of producing a test facility and antenna housings for an extended-range PIT tag transceiver system, the ultimate goal of the NMFS program is to interrogate adult salmon ascending fish ladders. To that end, procedures were explored and are presented to install, maintain, and remove extendedrange PIT tag transceivers in main ladders.

#### **AUTHORIZATION**

This Predesign report was prepared in accordance with the contract between the Department of Commerce, National Oceanic and Atmospheric Administration, National Marine Fisheries Service, Coastal Zone & Estuarine Studies Division, and Summit Technology dated February 17, 1995.



# INTRODUCTION

National Marine Fisheries Service (NMFS), with Bonneville Power Administration (BPA) support, is investigating the technical feasibility of using Passive Integrated Transponder (PIT) tags to monitor the movement of adult salmonids ascending fish ladders. This requires antennas and associated antenna housings to be designed, fabricated, and evaluated that are several times larger than presently used in the Columbia River Basin (CRB). In addition, these antennas must be of various geometries, completely waterproof, accessible for maintenance, and not alter the hydraulics of the fish ladder.

Two general categories of extended-range PIT tag antenna housings are required, passby and pass-through. The first approach would be used in connection with the VerticalSlot and Overfall-weir fish passage areas of a fish ladder. In the pass-by concept, PIT tag interrogation takes place while fish pass by a flat-plate antenna housing placed in a vertical or horizontal position. The second approach is used with orifices located within a fish ladder. Here fish being interrogated for PIT tags, pass through an opening within the antenna housing. In either case, the antenna housings must accommodate a range of antenna coil designs and be easily installed, removed, and adjusted for various testing conditions.

Once fabricated, the antenna housings, antennas, and associated PIT-tag transceiver electronics must be evaluated. To minimize impact and maximize flexibility in testing, NMFS sought a test fish ladder that could be altered to emulate existing fish ladders in the Columbia River Basin. The final selection of such a facility would be dependent upon agreement and cooperation from all stakeholders.

Working toward this goal, NMFS proposed to contract an independent structural engineering firm to:

Evaluate the technical feasibility of using Bonneville Dam's Fisheries Engineering Laboratory fish ladder to study PIT tag transceivers and antenna designs for the interrogation of adult fish.
Design PIT tag antenna housings that: a) enable a variety of antenna coil designs to be evaluated within a single housing, and b) can be deployed in an adult test

facility without altering the facility's structural integrity, hydraulics, or



modifying fish behavior.

FEBRUARY 1996



#### TEST FACILITY LOCATION AND DESCRIPTION

The first goal of this design effort was to evaluate the technical feasibility of using Bonneville Dam's Fisheries Engineering Laboratory (FEL) fish ladder to study PIT-tag transceivers and antenna designs for the interrogation of adult fish. This ladder is located adjacent to the North Shore Fish Ladder at the Bonneville Dam's 2ND Powerhouse. Plans, sections, and details of the existing North Shore Fish Ladder and Fisheries Engineering Laboratory fish ladders are shown in Figs. 1 through 7. The question as to the ladder's availability for such a program was not part of this effort.

The antenna housings to be evaluated at the facility are designed to emulate the VerticalSlot and Orifice/Weir portions of a fish ladder. The antenna housings thus were designed as replacement sections for the areas of interest. For the Vertical-Slot area, a sectional vertical flat-plate or pass-by antenna housing was designed. A similar pass-by antenna housing placed horizontally is suggested for the weir portion of the Orifice/Weir area, while a pass-through antenna housing is used with the orifice. Details of these housings are described elsewhere in this document.

For the FEL fish ladder to be considered as a PIT tag systems evaluation site, the ladder had to meet several criteria:

- 1. The weir and orifice geometries in the test ladder must be representative of the weir and orifice geometries in operating fish ladders within the CRB (e.g., North Shore Fish Ladder at the Bonneville 2ND Powerhouse).
- 2. The test ladder must be operable at various controllable water flow rates; it also must be able to be dewatered on demand.
- 3. The test ladder should be readily accessible by equipment needed to install and remove the antenna housings.
- 4. The test ladder must be capable of being altered without jeopardizing its integrity or functionality now or in the future.

The FEL's facility meets all of the above criteria.

At this time, it is understood that during the antenna evaluation period at the FEL fish ladder that both antenna areas, Vertical-Slot and Orifice/Weir, will not be operate simultaneously. All hydraulic calculations and conclusions are based on this assumption.

Figures showing the general arrangement and existing conditions at the proposed test ladder are provided at the end of this report. Fig. 1 shows the general arrangement plan of the North Shore features at Bonneville 2ND Powerhouse. Fig. 2 shows the Entrance and Exit Fish Ladders in plan view. Fig. 3 shows a plan detail of the Entrance Fish Ladder with the proposed locations for the test antennas highlighted. Partial sections of the Entrance Fish Ladder are shown in Fig. 4. Also in Fig. 4 the proposed locations for the test antennas are highlighted.



Figures showing existing conditions at a representative main ladder are also provided. These figures are provided for comparison to the proposed antenna housing drawings. This comparison illustrates that the test antenna housings should not have fish passage or hydraulic performance different from the existing North Shore Fish Ladder. Fig. 5 shows a plan view of the vertical slots. A test antenna housing is being proposed which emulates these vertical slots. The proposed vertical slot antenna housing is described in this report in the section titled "Vertical-Slot Antenna Housing" and is shown in engineering drawings (Figs. 15 through 19). Fig. 6 shows a plan view of the orifice/weir section of the North Shore Fish Ladder. Fig. 7 shows the details of the orifice and weir geometries of the existing North Shore ladders. A test antenna housing is being proposed which emulates these orifice/weir sections. The proposed orifice/weir antenna housings are described in this report under the title "Orifice/Weir Antenna Housing" and are shown in engineering drawings (Figs. 20 through 24).

Isometric renderings of the antenna housing installation in the Entrance Ladder to the FEL have been prepared to illustrate the relationship between the antenna housing and existing ladder construction. These are Figs. 8 through 14.

Fig. 8 shows a view of the antenna housing installation looking down and toward the southwest. Fig. 9 shows the antenna housings in the Entrance Ladder looking east from above Entrance Weir 43.

Figs. 10 and 11 are looking through the north wall of the Entrance Fish Ladder at the Vertical Slot antenna housing installation. In Fig. 10 the preferred Vertical Slot antenna housing is illustrated. Fig. 11 illustrates the alternate Vertical Slot antenna housing.

The downstream face of the Orifice/Weir antenna housing is shown in Fig. 12. Fig. 13 shows the upstream face of the Orifice/Weir antenna housing. The final isometric rendering is Fig. 14, showing an exploded view of the Orifice/Weir antenna housing and structural frame.



# **ANTENNA HOUSINGS**

The second task of this effort was to design antenna housings that: a) enable a variety of antenna coil designs to be evaluated within a single housing, and b) can be deployed in an adult test facility without altering the facility's structural integrity, hydraulics, or modifying fish behavior. The general location and approach taken for the housing are described in the previous section. A detailed description of the antenna housings designed for use in the Vertical-Slot and Orifice/Weir area of the fish ladder follows.

#### **VERTICAL SLOT ANTENNA HOUSING**

# Description

The Vertical-Slot antenna housing is patterned after Baffles No. 15, 16, and 17 of the Bonneville 2ND Powerhouse, North Shore Fish Ladder, Exit Control section. For the geometry of these orifices, see Fig. 5 (Vertical Slot Orifice Plan).

Two proposed designs have been prepared to model these orifices in the test ladder: a Preferred Vertical Slot antenna housing, and an Alternate Vertical Slot antenna housing. The Preferred Vertical Slot has been so designated because the geometry of its proposed installation allows greater flexibility in modeling the actual ladder hydraulic conditions. Of the two alternative designs prepared for consideration, only one will be selected and constructed.

The term Vertical-Slot antenna housing is used when characteristics common to both the Preferred and Alternate antenna housings are discussed in this section.

The Vertical-Slot antenna housing consists of a guide wall and the slot walls. The guide wall will be installed on the upstream side of the antenna housing. The slot walls consist of two portions: a fixed portion, and an adjustable portion. Both the guide wall and the slot walls will be fabricated of Series 500/525 structural fiberglass shapes and plates. All framing members will be 8-inch channels covered with 0.5-inch-thick fiberglass skin plate. Although the antenna housing is constructed primarily with adhesive-bonded connections, the design allows for bolted connections. This will permit the antenna housing to be modified independent of the unit's structural frame. The Preferred Vertical Slot antenna housing drawings are shown as Fig. 15 (Plan) and Fig. 16 (Elevations). The Alternate Vertical Slot antenna housing drawings are shown as Fig. 17 (Plan), Fig. 18 (Elevations), and Fig. 19 (Sections and Details).

Antenna housings will be enclosed in the downstream end of the fixed portion and the upstream end of the vertical slot's adjustable portion. The unused portion of the housing is currently shown on Fig. 17 to be filled with closed cell polystyrene foam. The unused portion could also be left as an air void. Filler materials will be selected based on their dielectric characteristics and their stability in a moist environment. To ensure negative buoyancy, water will be allowed to enter compartments of the antenna housing or portions of the antenna housing will be ballasted.



To install the antenna housings, they will be slid vertically down into compartments within the Vertical Slot antenna housing (Figs. 15 and 17). Because the antenna housings will be positively buoyant, it will be necessary to force them down into the compartments. A cover plate and retainer will be installed to secure the housings. For tests that utilize less than the full-height antennas, the unused portion will be replaced by an antenna-shaped spacer.

Although not shown on the sketches, there will be provisions to connect the antenna leads to the PIT tag detection system. If required, provisions for a metal RF shield would be included above the antenna.

#### **Preferred Location**

The guide wall of the Preferred Vertical Slot antenna housing will consist of a wall perpendicular to the test fish ladder's longitudinal axis. This wall will be fabricated in three, full-height sections.

The full-height sections will be bolted to the floor using inserts with a spacing of 3 inches. This configuration-two 2-foot sections, one 1-foot section, and bolt inserts on 3inch spacing-allows the fixed portion of be adjustable from 1 foot to 5 feet wide, and on either side of the test ladder.

This adjustability allows the tests to be conducted with arrangements which best initiate parallel flow lines through the antennas and minimize the potential for short-circuiting the flow.

The slot walls of the Preferred Vertical Slot antenna housing will consist of two portions, each containing an antenna housing oriented parallel to the other, forming the slot. These portions will be installed at a slope of 0.25:1.00 from perpendicular to the fish ladder's longitudinal axis. The portions will be bolted to the floor and each wall of the test ladder. The right-hand portion of the slot wall (looking downstream) will be bolted in a fixed location. The left-hand section provides adjustability.

A false floor will be installed to raise the floor elevation to 42.0 feet. This floor will be sloped on the downstream face to minimize the locations where fish could hold up.

Provisions will be made for a partial-height, adjustable, vertical barrier on the downstream edge of the vertical slot. This vertical barrier has been proposed to facilitate testing of the antennas. With the barrier removed, full-depth tests can be run to prove antenna performance at hydraulic pressure heads of up to 12 feet. The vertical barrier can then be installed and tests run at varying flow velocities. The velocity of the water through the slot is dependent upon the height and placement of the vertical barrier.

In the North Shore Fish Ladder, Exit Control section, feet-per-second (fps) slot velocities, including carryover, have been observed to be approximately 3.5 fps to 6.0 fps. The FEL Entrance Fish Ladder design flow is 33 cubic feet per second (cfs). To representatively model slot velocity and the ladder flow rate concurrently, it will be necessary to adjust the slot's width and the vertical barrier's height.



#### **Alternate Location**

The guide wall of the Alternate Vertical Slot antenna housing also will consist of a wall perpendicular to the longitudinal axis of the fish ladder. As currently proposed, the guide wall is joined to the fixed portion of the slot walls.

The fixed portion of the slot walls will consist of a stem wall extending downstream at a slope of 0.25:1.00 from parallel to the longitudinal axis of the fish ladder. This portion will be fabricated in one section; the panel is then bolted to the floor of the fish ladder.

The adjustable portion o tl~,e st alls will also be installed at a slope of 0.25:1.00 from parallel to the longitudinal axis of the fish ladder, constructed in one section, and bolted to the floor. The bolts will be provided with a spacing of 3 inches. This spacing, along with oversized holes in the adjustable portion, will allow the vertical slot to be set at openings of 1 foot to 2 feet, in 3-inch increments. One upstream and one downstream closure plate will be provided for each opening increment, for a total of five sets of plates.

A false floor will be installed to preclude the fish avoiding detection by swimming below the antennas. The false floor is sloped on the downstream face to help minimize the tendency for fish to hold up in front of the raised floor.

#### Installation

Vertical Slot antenna housings have been designed for two locations in the proposed test ladder. While only one will be installed, both alternates would be installed in a similar manner, by bolting to the existing walls and floor. Since installation of the Vertical Slot antenna housing will not require removal of concrete portions of the existing fish ladder, no concrete replacement will be required.

## **Preferred Location**

The selected location for the Preferred Vertical Slot antenna housing is in the large pool upstream of Weir 53. (Refer to Fig. 3 for location.) This pool, the first downstream from the FEL, is 8 feet wide by approximately 26 feet long.

The large pool area allows flexibility in the alignment and placement of the antenna housing. This flexibility allows a design that models the water flow patterns of the North Shore Ladder, Exit Control section. It is this flow modeling which makes the large pool upstream of Weir 53 the preferred alternative location.

In the selected pool, the floor is level at El. 38.00, and there is no sill, orifice, or weir adjacent to the proposed antenna housing location.

#### Alternate Location

The selected ladder location for the Alternate Vertical Slot antenna housing is between Weir 51 and Weir 52 in the FEL Entrance Fish Ladder (Fig. 3). Each section of the fish ladder from Weir 46 through Weir 52 has a level floor with a sill at each weir which is 2.5 feet high. An orifice is formed in each sill. The top of the sill at Weir 52 is El. 40.50.



Installation of the adjustable portion of the Vertical Slot antenna housing will require that the weir gate and its appurtenances at Weir 51 are removed, and that the orifice in the sill is covered. Because of the number of anchor bolts to be installed, and the critical nature of their location, it is expected that the fiberglass fabricator will prepare a template to match the drilling of the members.

# Operation

Both Vertical Slot antenna housings operate in similar fashion. 'The tests can be run at various flow rates, flow depths, and slot velocities that approximate that of the main fish ladder.

The proposed test ladder design flow is approximately 33 cfs. Supply flow is comprised of water diverted from the main ladder and water supplied by a floor diffuser in the FEL. The supply diffuser in the FEL has a maximum capacity of 60 cfs. For high-flowrate tests of the Vertical Slot antenna housings, it may be necessary to divert excess flow from the test ladder before it is discharged to (and disrupts flow in) the main ladder. This flow could be diverted by temporary pumping or by the installation of a screened and gated pipe that discharges into the existing storm drain manhole.

# **Preferred Location**

The Preferred Vertical Slot antenna housing will be designed to operate with a maximum water depth of 13 feet and with a maximum width of 2 feet. The antenna housing would be operated to provide a range of test velocities and flow depths by varying the flow rate, slot area, and number of downstream weir gates open.

The slot area may be varied not only by adjusting the slot width, but by adjusting the height also. As a result, the partial-height, adjustable, vertical barrier is proposed. With a 2-foot slot width and a ladder flow rate of 35 cfs, all but approximately 3 to 4 feet of the slot would need to be blocked off to produce an average velocity of 6.O fps. This velocity, although representative of existing slot flow velocity, is perhaps somewhat excessive. However, the use of blocking to increase slot velocities could be used to some degree, with the open portion of the slot positioned at any desired elevation.

At full design depth and maximum slot width, the flow through the slot at a 1.0-foot drop will be approximately 144 cfs. For a slot width of 1.5 feet, the flow is approximately 108 cfs; for a slot width of 1.0 feet, the flow is approximately 72 cfs.

#### Alternate Location

The Vertical Slot antenna housing will be designed to operate with a maximum water depth of 13 feet and a width of 2 feet. This test would be operated to provide a range of test velocities by varying the ladder flow rate and the slot width. Additional operational flexibility is achieved by opening or closing the downstream weir gates.

The water surface upstream of Weir 52 will be at El. 52.50. With a 1-foot drop, the flow through a 2.0-foot slot will be approximately 144 cfs. For a slot 1.5 feet wide, the flow is approximately 108 cfs. For a slot 1.0 feet wide, the flow is approximately 72 cfs.



#### Performance

The Vertical Slot antenna housing is patterned after Baffles No. 15, 16, and 17 of the Bonneville 2ND Powerhouse, North Shore Fish Ladder, Exit Control Section. For the geometry of existing orifices, see Fig. 5 (Vertical Slot Orifice Plan). The existing slot is 1.969 feet wide in that set of baffles. The antenna housing opening will be nominally 2.0 feet wide and has approximately 0.375-inch adjustability. The antenna housing will have an adjustable opening width which will allow the modeling of additional sets of baffle. For example, Baffles No. 16 and 17, with an opening of 1.719 feet, could be modeled by setting the antenna housing at 1.75 feet and taking up all the slack in the hold-down bolts.

Both the antenna housing slot and the existing ladder slot are sloped 0.25:1.0 with respect to the longitudinal axis of the fish ladder.

With similar opening width, opening height, and angle to flow it is anticipated that the antenna housing will have fish passage and hydraulic performance similar to the existing North Shore Fish Ladder.

#### Removal

Upon completion of the tests, the anchor bolt sleeves installed in the existing ladder will have plugs installed and left flush.

#### **ORIFICE /WEIR ANTENNA HOUSING**

# Description

The Orifice/Weir antenna housing will be constructed as a full-scale model, or prototype, of Entrance Weirs 38 through 43 and Exit Weirs 44 through 48 of the FEL fish ladder (see Figs. 1, 2, 3, and 7). Conceptual designs have been prepared for two Orifice/Weir antenna housings: a Structural Fiberglass Orifice/Weir antenna housing constructed of fiberglass shapes and plates, and a Fiberglass Reinforced Concrete antenna housing. Of these two designs only one will be built.

Two types of antenna will be tested in the Orifice/Weir unit. A flat antenna will be tested in the weir section, and a loop antenna will be tested in the orifice section. The antenna housing will be removable for modification or maintenance. If necessary, provisions for a metal RF shield will be made above the antenna housing to provide RF shielding.

# Structural Fiberglass

A Structural Fiberglass Orifice/Weir antenna housing will be constructed of Series 500/525 structural shapes and plates. For the upstream elevation and sections pertaining to this antenna housing, see Fig. 20 (Elevation and Section) and Fig. 21 (Sections).



The frame will consist of 6-inch channel sections and will be covered with 0.5-inch-thick plate. This results in a total thickness of 7 inches for the structure. This is 1 inch less than the current Weir 42.

The antenna housing will be 8 inches thick and will have the upstream and downstream chamfer profiles similar to existing weirs. The unit's structural frame is constructed with adhesive bonded connections. Design of the antenna housings allows for removable and interchangeable antenna sections, which permits the antennas to be modified

independent of the unit's structural frame. The antennas are enclosed in annular portions of the antenna housing. This housing is currently shown to be filled with closed cell polystyrene foam or left as an air void. Filler materials will be selected based on their dielectric characteristics and their stability in a moist environment. To ensure negative buoyancy, water will be allowed to enter cells of the antenna housing or portions of the antenna housing will be ballasted.

Proposed construction of structural frames for the antenna housings is of structural fiberglass plates and shapes, with standard colors of olive green and haze gray. It is anticipated that these colors will not distract the fish and, therefore, will not disrupt fish passage. Proposed construction of antenna housings is of sheets of Lexan® plastic, which may be painted to match the structural frames.

Although not shown on the sketches, there will be provisions to connect the antenna leads to the RF generator and data collection system.

# Fiberglass Reinforced Concrete

A fiberglass-reinforced, concrete Orifice/Weir antenna housing would be constructed using moderate-strength, 5,000-pounds-per-square-inch (psi), portland cement concrete. Structural reinforcing will be Thermal Cure® fiberglass rod and bar. Temperature reinforcing will be synthetic fiber reinforcement.

The antenna housing will have thickness, geometry, and surface material identical to the existing Weir 42. Antennas will be enclosed in housings fabricated of Lexan®, and will be removable. The profile of the overflow weir and the orifice chamfers will match existing profiles. For construction details of the antenna housing, see Fig. 22 (Elevation and Section) and Fig. 23 (Sections).

Although the sketches of this antenna housing show an unreinforced section, and preliminary design calculations indicate that lower strength concrete (as low as 1,000 psi) may be used without reinforcement, it is not recommended that the antenna housing be constructed without structural and temperature reinforcement. The concrete strength and reinforcing density will depend on the final design criteria selected and the desired "toughness" of the unit. For all selections of design criteria, concrete strength, and structural reinforcing density it is recommended that fibrous reinforcement be used for shrinkage and temperature crack control, and that fiberglass rod be used as structural reinforcement. Additionally, it is recommended that concrete used have a design compressive strength of 5,000 psi.



The antennas are shown as being enclosed in a housing, fabricated of Lexan®, similar to the Structural Fiberglass Orifice/Weir antenna housing. The housing will be filled with the same materials as the other antenna housings.

Although not shown on the sketches, there will be provisions to connect the antenna leads to the PIT tag detection system.

#### Installation

It is proposed that the orifice/weir antenna housing be installed at the Weir 421ocation in the FEL Entrance Fish Ladder (Fig. 3). This location was selected because it is in a location of established flow and is accessible. Weir 42 is nearly the middle weir on the south side of the ladder, is in the sloping floor portion of the ladder, and is bordered by fixed overflow weirs both upstream and downstream. (Weirs 44 through 53 have adjustable gates; Weirs 38 through 43 are fixed overflow sections.) Additionally, Weir 42 is not too deep in the ladder channel, and it does not have a strut above it as do Weirs 39 and 40. The only other weir to meet the same criteria, Weir 41, is in an equivalent location; however, Weir 42 was selected.

Removal of the existing overflow portion of the weir would be required, and this would entail cutting the concrete and reinforcing steel in the wall. In addition, a portion of the concrete beneath the weir must be removed to accommodate the loop antenna around the orifice at the weir bottom.

Construction drawings for the ladder specify a minimum cover of 4 inches for unformed surfaces. This should minimize the need to cut any reinforcing steel in the floor of the ladder.



## Operation

A removable antenna housing will be installed as the top portion of the overflow weir. Another removable antenna housing, installed in the structural frame, will serve as the orifice. As fish swim through the orifice or over the weir, the PIT tag will be energized by an electromagnetic field originating in the antennas. The PIT tag then discharges and the signal is received by the antenna. Excess RF radiation will be trapped by the RF shield provided above and beside the antenna units.

### **Performance**

Fish passage and hydraulic performance of the antenna housing should be similar to the existing weir/orifices in the FEL Entrance Fish Ladder and the North Shore Fish Ladder. Similar performance will be achieved with this antenna housing by fabricating it to the same geometry as the present Weir 42 of the FEL Entrance Fish Ladder. Weir 42 of the FEL Entrance Fish Ladder is hydraulically similar to one side of the weirs in the North Shore Fish Ladder of the 2ND Powerhouse. Consequently, the prototype installed in place of Weir 42 should also perform similarly to the weirs in the North

Shore Fish Ladder of the 2ND Powerhouse. The geometry of Weir 42 is shown in Fig. 7 (Orifice and Weir, Details).

#### Removal

Upon completion of the test program, the Orifice/Weir antenna housings will be removed from the Entrance Weir. The fiberglass anchor bolts will be cut off and reinforcing steel will be doweled into the walls and floor. The dowels will be similar to those cut out during the installation of the antenna housing. They will be offset to avoid the cut-out reinforcing. Once the dowels are installed, the orifice/weir concrete will be replaced as shown in the original construction drawings. Any temporary bolt holes will be grouted flush.



## RADIO FREQUENCY (RF) SHIELD

The PIT tag antenna is both an RF transmitter and receiver. To reduce the release of stray RF emissions, provisions for an RF shield will be made above the antenna housing and the adjacent bays of the ladder. One possible design is shown in Fig. 24 (Radio Frequency Shield - Plan and Section).

The RF shield would be configured as a modular, removable access platform with grounding capability. There would be three 8.0-foot panels marked Left, Center, and Right. The Left and Right panels would have a guardrail on one end; the center panel would not. This modular layout would allow the shield to cover beyond two pools of the ladder, or, with the center panel omitted, to cover only the antenna housing. If it is found necessary to extend the RF shield, additional center type panels could be fabricated and installed. In addition to the horizontal shielding provided by the grating, there would be adjustable wire fabric on the ends of the Left and Right panels to capture RF emissions traveling with a horizontal component.





## **APPLICATION TO MAIN LADDER OPERATION**

#### **OVERVIEW**

This predesign report addresses the feasibility of producing a test facility for an extended-range PIT tag interrogation system. However, the goal of the NMFS program is to interrogate adult salmon in ascending fish ladders. To this end, procedures were investigated to install, maintain, and remove the extended-range PIT tag monitors in main ladders.

This section of the report will identify the concerns raised in translating the extendedrange PIT tag monitoring equipment from a test ladder to an operating main ladder. It will also propose ways in which this translation may be accomplished.

#### INSTALLATION

Installation in an operating ladder will be more constrained than installation in the test ladder. Initial installation will be limited to the period of scheduled ladder shutdown, and in-service repairs will need to be accomplished without dewatering the ladder.

Initial installation of the systems will be accomplished during the in-water work period at the projects, which is typically during the winter months. The time required for modifications to the ladders is relatively modest and can be accomplished during this time period. Preliminary testing with water flowing in the ladder should also be accomplished during the in-water work period, so that the ladder can be dewaeered if necessary.

At the end of the in-water work period, any maintenance necessary to the units will need to be accomplished without dewatering the ladder. It may be necessary to change antennas during operation of the ladder.

In the vertical slot monitor, changing the antennas can be accomplished with no disruption to ladder operation. The antenna housing units can be withdrawn from the top of the frames and modified, rebuilt, or replaced as necessary.

Changing antennas in the orifice/weir units is more difficult. It will be necessary to remove the weir panel out of the fishway, replace the antenna housing with a spare, and return the fish ladder to normal operation. The antenna housing can then be taken to a shop where the antenna housings are disassembled and the antennas are rebuilt.

In order to remove the weir panel, it will be necessary to stop the flow over and through the weir. Since the typical ladders have two overflow weirs, all of the flow can be routed to one of them while the other is blocked off. Although this flow routing will have a substantial effect on the local hydraulics of the ladder, it will occur for a relatively short period of time and will not block fish passage.



As indicated on Fig. 25 (Stoplogs - Plan and Section), the stoplogs will consist of two

One section is a 0.25-inch-thick steel plate with angle stiffeners which will be installed parallel to the flow in the ladder. This section will be installed first, with water pressure holding the downstream edge against the return wall of a weir. The upstream end of the section is braced off the exterior wall of the ladder with double angle struts.
 The other section of the stoplogs is a 0.25-inch-thick steel plate with nylon skids. The skids will consist of two sets of MC901 nylon strips. One set of strips, each strip approximately 2 inches wide, will be attached to the face of the stoplog section. The other set of strips will be installed on the face of the existing

concrete weir wall. The sets of strips will be placed such that the stop log and wall make contact, nylon to nylon. The skids will decrease frictional resistance to sliding of the section against the upstream face of the weir and orifice wall.

A stoplog will be installed in the affected side of the ladder. This will cause the water level to rise approximately 0.8 feet, which will cause the weir divider wall to overtop slightly. Two options are available to reduce the water level upstream of the stoplog.

The first option is to adjust the existing control section of the ladder to temporarily reduce the flow. This flow reduction is on the order of 32 percent and should occur only for a period of two to four hours.

The second option is to install an operable slide gate in the area between the two weirs. Before installing the stoplogs, the gate would be opened to reduce the differential at the affected weir.



## **FIGURES**

#### **ORGANIZATION**

The figures for this report consist of drawings and renderings. They have been divided into three groups.

The first group of figures (Figs. 1-7) are extracts from the construction drawing set for the Bonneville 2ND Powerhouse, U.S. Army Corps of Engineers, Portland District. These seven figures are intended to illustrate the general arrangement and existing conditions at the proposed test ladder.

The second group of figures (Figs. 8-14) are isometric renderings of the antenna housings installed in the Entrance Ladder to the FEL. (Note that vibrant colors are used to highlight the antenna housings and help distinguish them from the existing structure.) This group of seven figures is intended to provide an artist's rendering of the proposed installation and to allow the viewer to see that the fish passages will not be altered by installation of the proposed antenna housings.

The third group of figures (Figs. 15-25) include conceptual design drawings of the antenna housings. This group of 11 figures is intended to provide the detailed dimensions which allow verification that weir and orifice surfaces of the antenna housings match the analogous surfaces on the existing structure. In addition, these figures allow the test equipment's structural integrity to be determined.

### **TITLE LIST**

FIG.	TITLE
Construction Drawing Set-Bonneville 2ND Powerhouse (selected)	
1 2 3 4 5 6 7	Shore Features - Plan Fisheries Engineering Laboratory - Entrance and Exit Fish Ladders, Plan Fisheries Engineering Laboratory - Entrance Fish Ladder, Plan Fisheries Engineering Laboratory - Entrance Fish Ladder, Sections North Shore Fish Ladder - Vertical Slot Orifice, Plan North Shore Fish Ladder - Orifice/ Weir, Plan Existing Ladders - Orifice and Weir, Details
Isometrio	c Renderings
8 9	Antenna Housing Installation - Looking North from the Southwest Antenna Housing Installation - Looking Southwest from above Weir 43

Antenna Housing Installation - Looking through Wall into Poo153 from Southeast



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- Antenna Housing Installation Looking through Wall into Pool 51 from Southeast
- 12 Antenna Housing Installation Looking North from Just Downstream of Weir 42
- Antenna Housing Installation Looking Southwest from Just Upstream of Weir 42
- 14 Weir Slot Removal at Weir 42, Exploded View of Orifice/Weir Antenna Housing

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- 17 Vertical Slot Antenna Housing (Alternate Location) Plan
- 18 Vertical Slot Antenna Housing (Alternate Location) Elevations
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- 22 Orifice/Weir Antenna Housing Elevation and Section (Fiberglass Reinforced Concrete)
- 23 Orifice/ Weir Antenna Housing Sections (Fiberglass Reinforced Concrete)
- 24 Radio Frequency Shield Plan and Section
- 25 Stoplogs Plan and Section



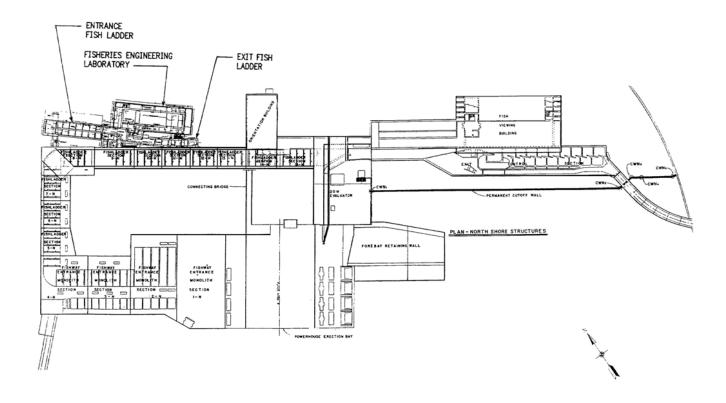


Figure 1. Shore Features - Plan



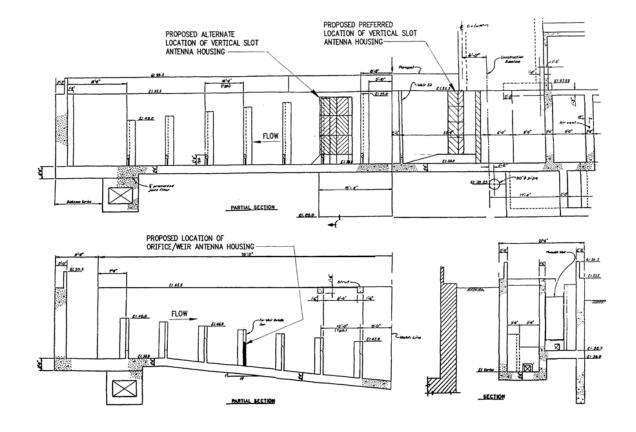


Figure 2. Fisheries Engineering Laboratory - Entrance and Exit Fish Ladders, Plan



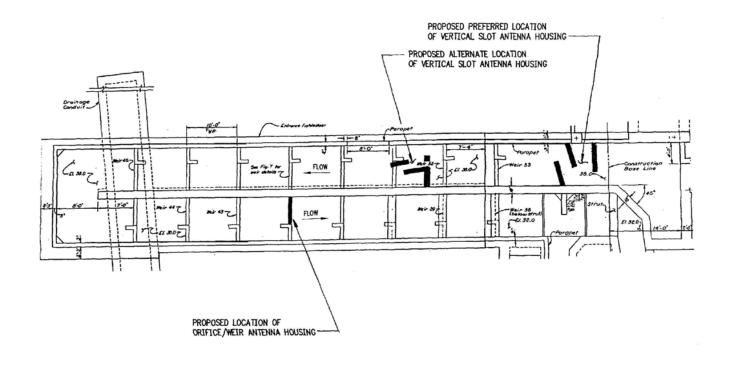


Figure 3. Fisheries Engineering Laboratory - Entrance Fish Ladder, Plan



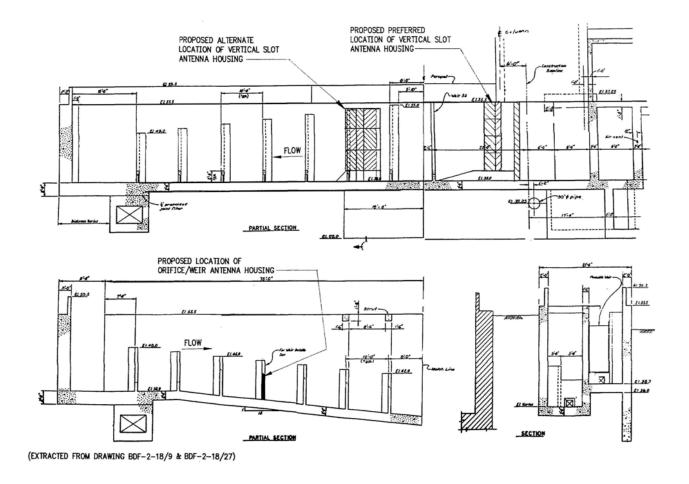


Figure 4. Fisheries Engineering Laboratory - Entrance Fish Ladder, Sections



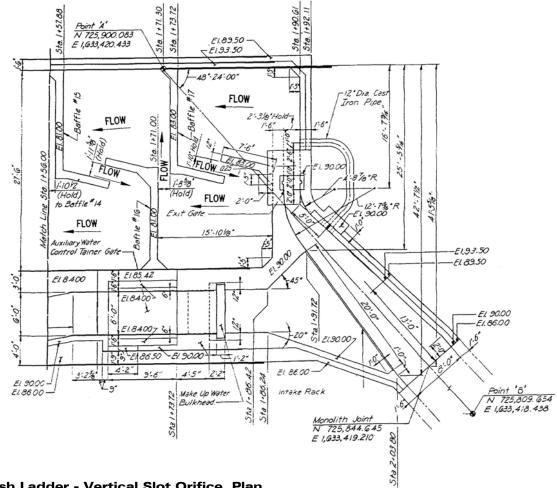
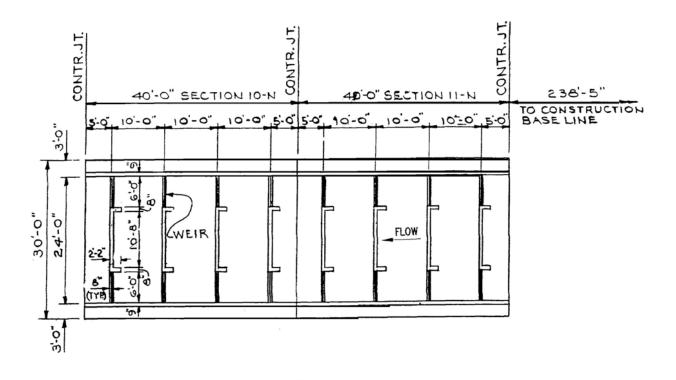


Figure 5. North Shore Fish Ladder - Vertical Slot Orifice, Plan





PLAN-EL.58.5 3/32"\* 1'-0"

Figure 6. North Shore Fish Ladder - Orifice/ Weir, Plan



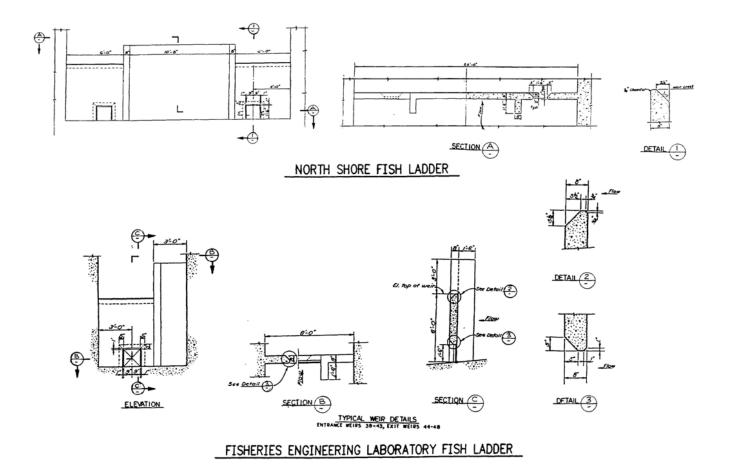


Figure 7. Existing Ladders - Orifice and Weir, Details



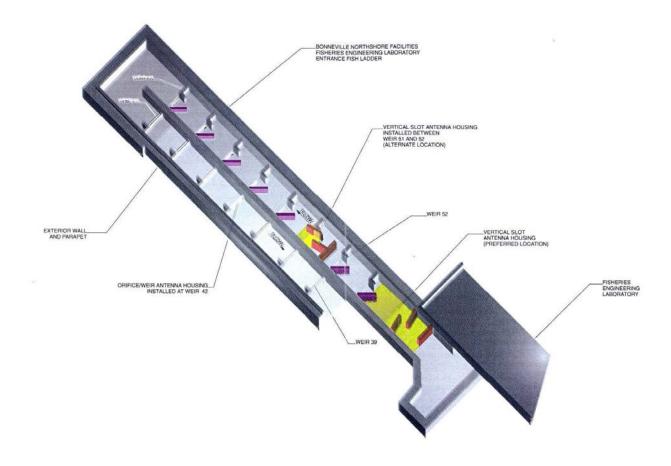


Figure 8. Antenna Housing Installation - Looking North from the Southwest



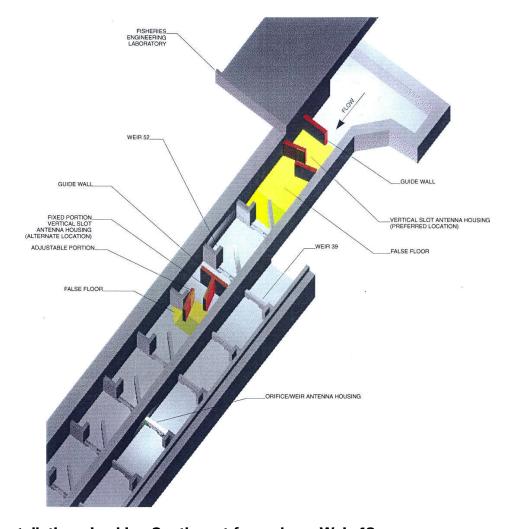


Figure 9. Antenna Housing Installation - Looking Southwest from above Weir 43



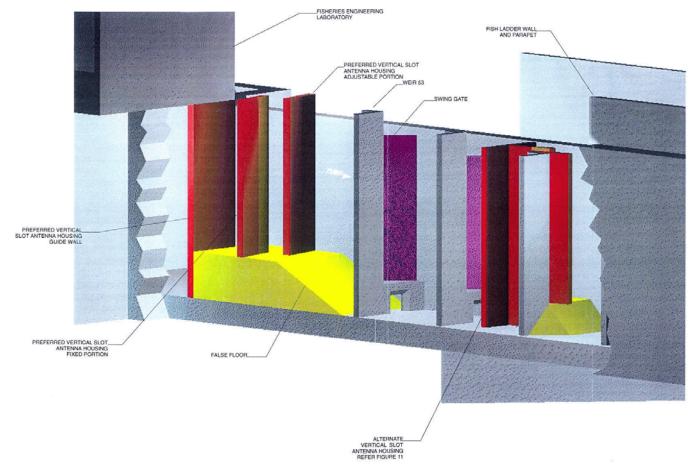


Figure 10. Antenna Housing Installation - Looking through Wall into Poo153 from Southeast



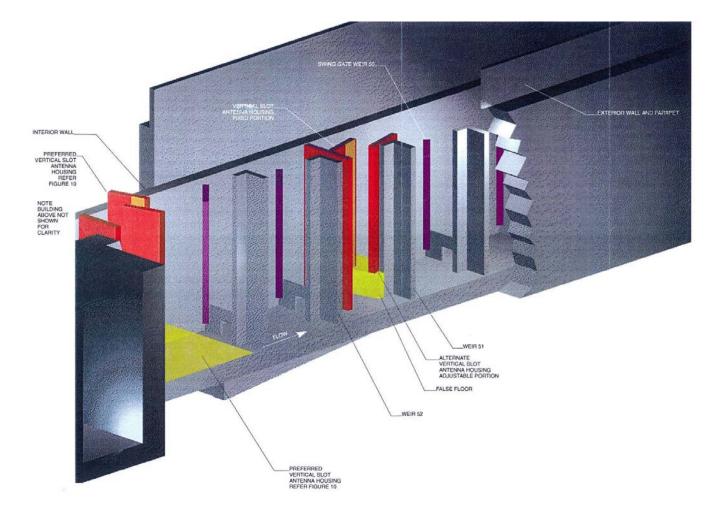


Figure 11. Antenna Housing Installation - Looking through Wall into Pool 51 from Southeast



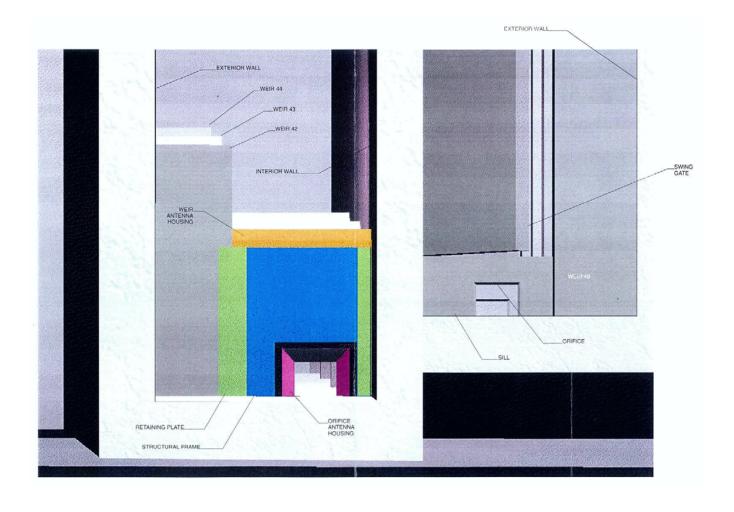


Figure 12. Antenna Housing Installation - Looking North from Just Downstream of Weir 42



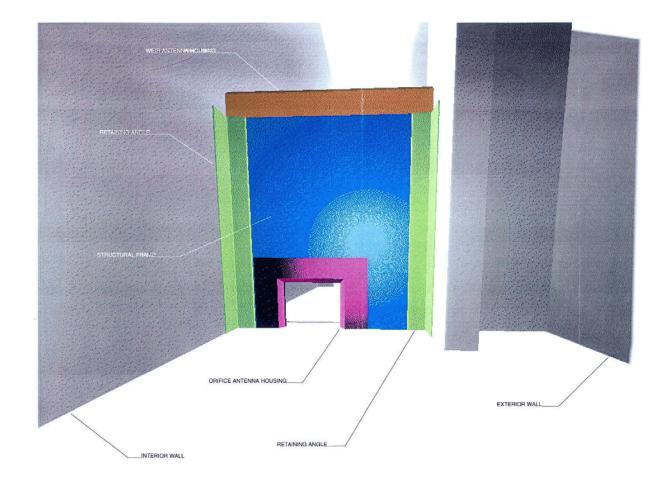


Figure 13. Antenna Housing Installation - Looking Southwest from Just Upstream of Weir 42



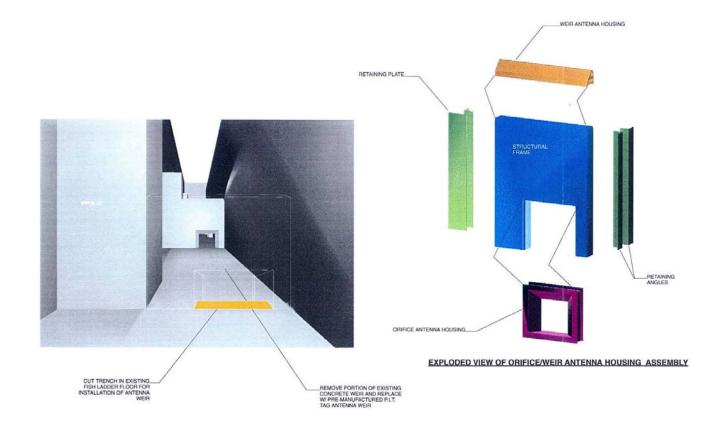


Figure 14. Weir Slot Removal at Weir 42, Exploded View of Orifice/Weir Antenna Housing



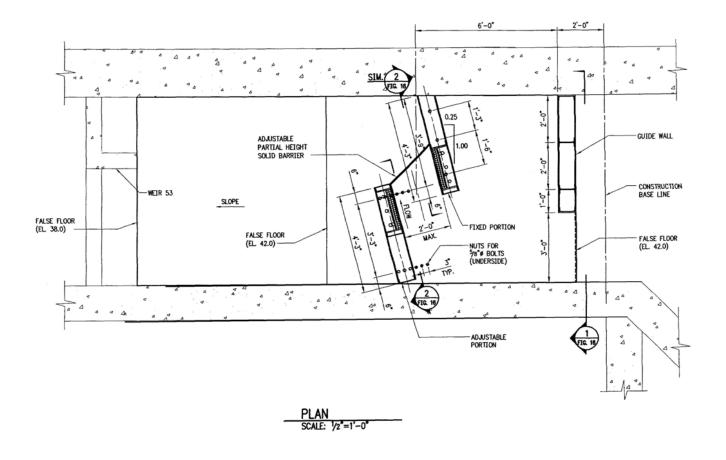


Figure 15. Vertical Slot Antenna Housing - (Preferred Location) - Plan



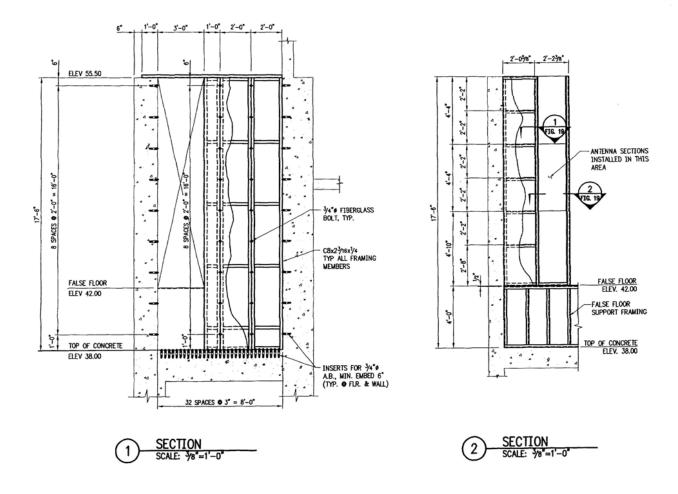


Figure 16. Vertical Slot Antenna Housing - (Preferred Location) - Sections



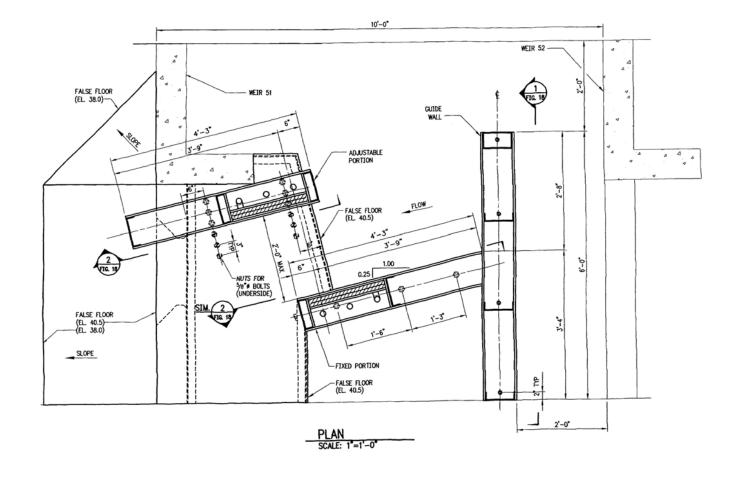


Figure 17. Vertical Slot Antenna Housing - (Alternate Location) - Plan



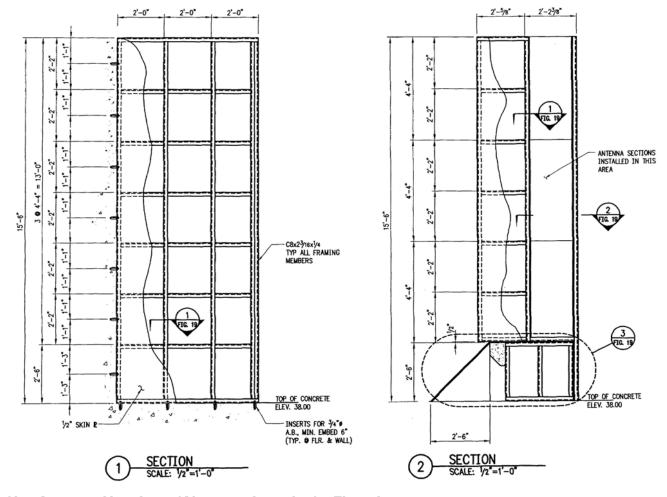


Figure 18. Vertical Slot Antenna Housing - (Alternate Location) - Elevations



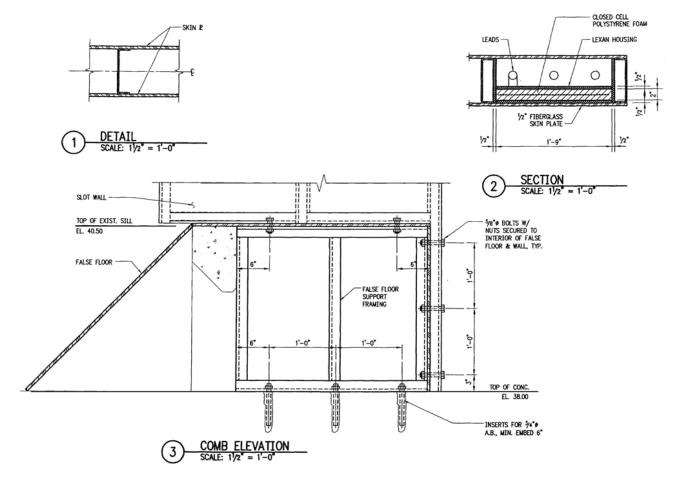


Figure 19. Vertical Slot Antenna Housing - (Alternate Location) - Sections and Details



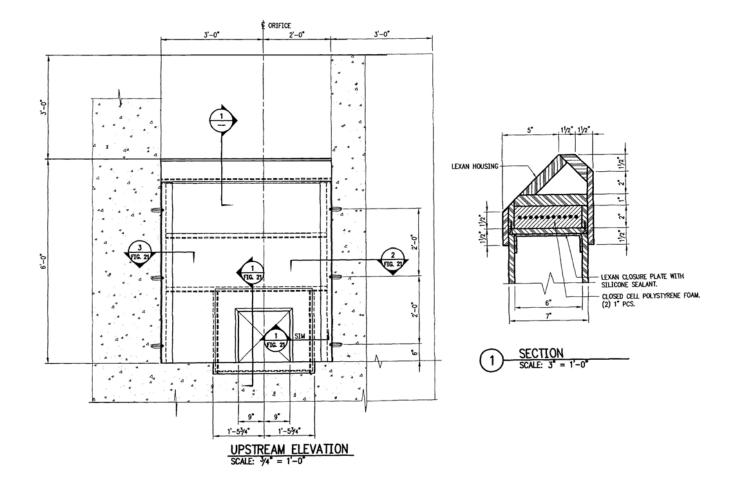


Figure 20. Orifice/ Weir Antenna Housing - Elevation and Section (Structural Fiberglass)



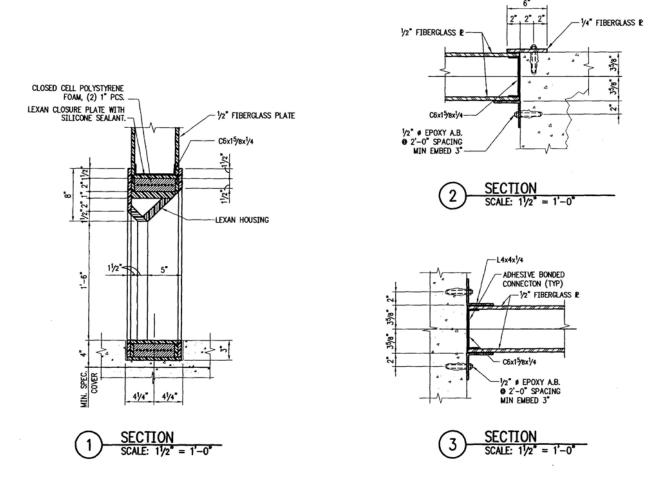


Figure 21. Orifice/Weir Antenna Housing - Sections (Structural Fiberglass)



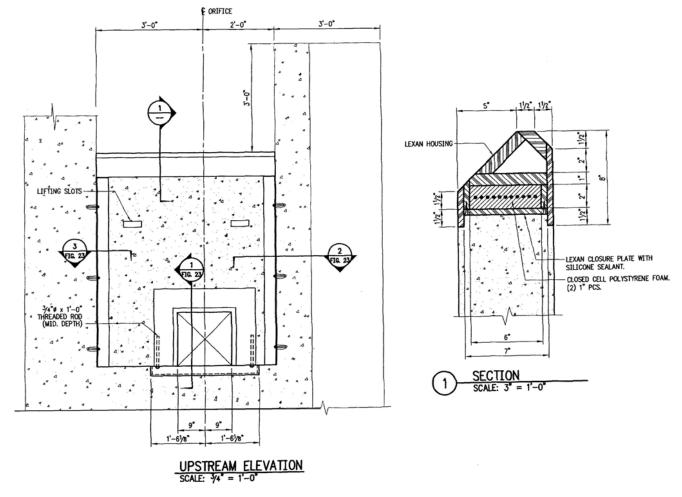


Figure 22. Oifice/Weir Antenna Housing - Elevation and Section (Fiberglass Reinforced Concrete)



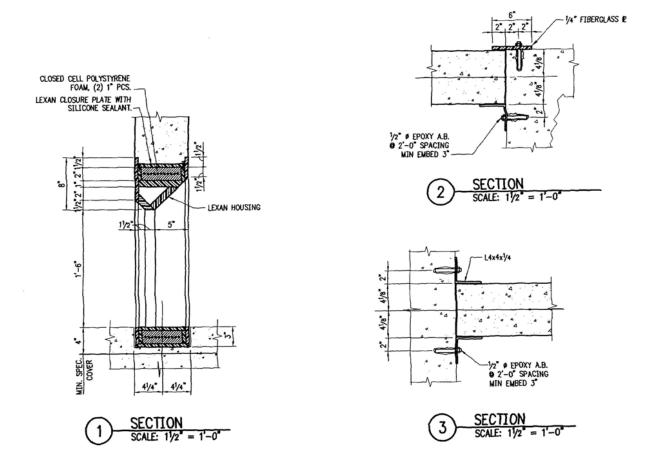


Figure 23. Orifice/ Weir Antenna Housing - Sections (Fiberglass Reinforced Concrete)

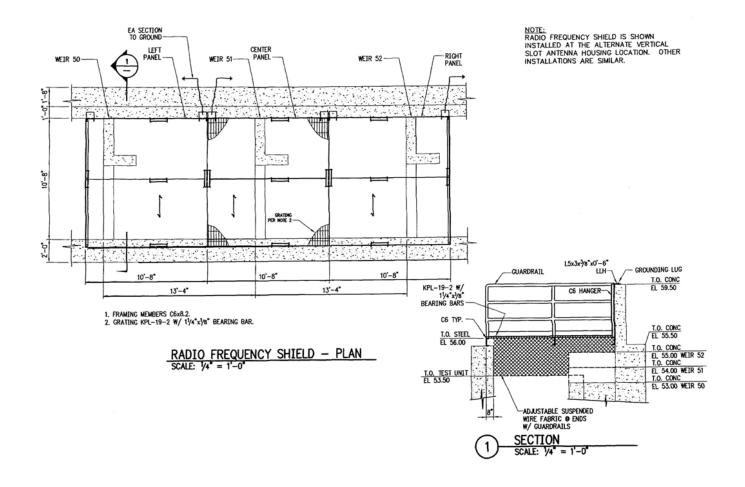


Figure 24. Radio Frequency Shield - Plan and Section

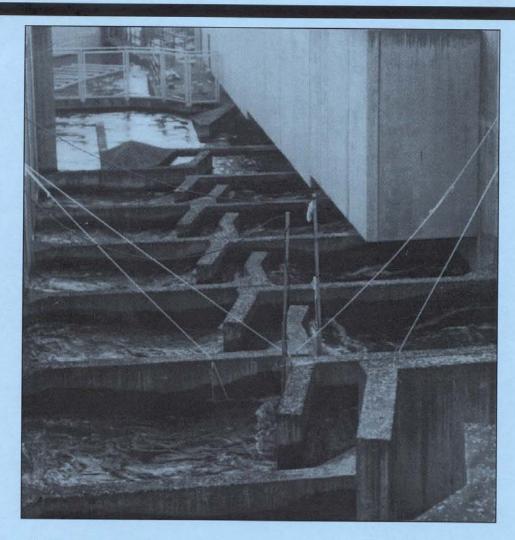


# **APPENDIX C**

# Fish behavior at Submerged Orifices. Overflow Weirs, and Vertical Slots in the Fish Ladder at Bonneville Second Powerhouse 1993-1994

Larry M. Beck U.S. Army Corps of Engineers





# **Fish Behavior**

at Submerged Orifices, Overflow Weirs, and Vertical Slots in the Fish Ladder at Bonneville Second Powerhouse 1993 - 1994

June 1995

# Fish Behavior at Submerged Orifices, Overflow Weirs, and Vertical Slots in the Fish Ladder at Bonneville Second Powerhouse 1993 - 1994

by

Larry M. Beck

U.S. Army Corps of Engineers CENPP-OP-SRF

Bonneville Lock and Dam Cascade Locks, OR. 97014 (541) 374-8801 March 14, 2003

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# Introduction

The National Marines Fisheries Service is developing passive integrated transponder (PIT) tag methodology to evaluate adult salmonid passage through hydroelectric projects on the Columbia River. PIT tags permit the identification of individual fish without handling after the initial marking, thus reducing stress. The best locations for PIT tag interrogators to monitor adult salmon passage at dams have not been determined. A concern regarding PIT tag interrogation systems is that they emit a strong electromagnetic field. This field may create a health risk for organisms with long exposure times (Earl Prentice personal communications,1993, NMFS, Manchester, Washington). Three potential locations for the PIT tag interrogation system to monitor adult salmon passage were identified: vertical slots, submerged orifices, and the overflow weirs in fish ladders.

Many studies have been conducted on fishways to estimate the rate of fish passage. These studies examined the time fish took to pass through different types of ladders, but not the length of time they stayed at any particular location. Elling and Raymond (1956), Long (1959), Gauley (1960), Bell (1962), Weaver (1962), and Gauley and Thompson (1963) showed median passage times ranging from 20 seconds (s) to 5.8 minutes (min) per pool for a weir and pool type ladder. Monan and Liscom (1974) showed a total passage rate of 15 min through the vertical slot section of Bonneville Dam's Bradford Island ladder. Monk et al. (1989) gave median passage times, ranging from 1.2 to 7.3 min per pool through a vertical slot ladder. Fish ladders were designed to provide resting areas (eddy and low velocity areas) and passage routes (high velocity areas). Passage routes between each pool are submerged orifices, overflow sections of weirs, and vertical slots. Most of the time that a fish spends at each pool is expected to be in the low velocity areas, resting. If the fish is in the low velocity areas most of time, the high velocity areas would be the better locations for the PIT tag interrogation systems because fish would be exposed to the electromagnetic field for the shorter time. To provide baseline data and design criteria, video cameras were used to determine behavior at the overflow sections, submerged orifices, and vertical slots.

## **Objectives**

The objectives were to document the following behaviors of fish in specified regions of a submerged orifice, a vertical slot, and an overflow weir. This information would be used to design PIT tag interrogation systems for adult fish.

- 1. Establish a baseline of fish behavior prior to installation of PIT tag monitors in the region of interest.
  - a. Types of fish movement.
  - b. Vertical distribution of fish.
- 2. Establish the amount of time available to interrogate fish.
  - a. Determine the amount of time a fish spends in the region.
  - b. Determine the fish's velocity through the region.

#### **Methods**

# Site Description

Bonneville Dam is the first dam upriver from the Pacific Ocean on the Columbia River, 64.4 km east of Portland, Oregon, at river km 234 (Figure 1). The Washington shore ladder is located on the north side of the second powerhouse (Figure 2).

Fish enter the adult passage facilities through the fish collection system (a system of floating orifices, large entrances, and channels). Fish proceed into a 7.32-m wide pool and overflow weir type ladder. Each weir is named by the elevation, in feet, of the crest of the overflow section. In addition, each weir has two 46-cm square submerged orifices on the floor of the ladder, each located 122 cm from the ladder wall, and two 152-cm wide overflow sections 183 cm from the ladder floor next to each wall (Figure 3). After the ladder, fish enter a junction pool where they join fish from the Cascades Island entrance. Fish are then crowded to facilitate counting. After passing the count station, fish pass into a section of pools (for this study, the numbering for pools at the vertical slots started at 1 at the count station) and vertical slots (Figure 4). After the section of pools and vertical slots, the fish enter a channel which exits into the forebay.

# Sampling Locations and Equipment

The submerged orifices of weir 65 and weir 66, were observed with two Simrad Osprey cameras<sup>1</sup>, model OE1359, rated at 0.03 lux with a 90° diagonal field of view. The cameras were 0.53 cm in diameter and 1.52 cm in length. One camera was placed facing weir 65 and the other camera facing weir 66 (Figure 5).

A frame, 172-cm wide by 276-cm long by 274-cm tall, was used to secure the cameras in the ladder for observing submerged orifices. This procedure allowed deployment without dewatering or modifying the ladder. A 46-cm wide, white metal plate attached across the bottom at each end of the frame added contrast to the floor of the fishway for the camera. On the plate were lines that were 15.2 cm apart, parallel to the flow. The cameras were placed in three locations designated as stations. The locations were:

- Station 1. One camera was located 32 cm upstream from the weir 65 south orifice, 158 cm from the wall, and 20 cm off the fishway floor. The second camera was located 36 cm downstream from the weir 66 south orifice, 158 cm from the wall and 20 cm from the fishway floor.
- Station 2. Bubbles hindered the view at station 1, so both cameras were moved 28.5 cm farther away from the orifices. One camera was located 56 cm upstream from the orifice, 158 cm from the wall, and 20 cm from the fishway floor. The second camera was located 58 cm downstream of the orifice, 158 cm from the wall, and 20 cm from the fishway floor (Figures 3 & 5).
- 1. The use of trade names does not imply endorsement by the government.

Figure 1. Columbia River Drainage.

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**OREGON** 

WASHINGTON

Figure 2. View of Bonneville Dam.

**Bonneville** 

Station 3. Both cameras were moved to observe fish behavior between the orifice and the ladder wall (Figures 3 & 5). One camera was located 91 cm upstream from the orifice, 13 cm from the wall, and 25 cm from the fishway floor. The second camera was located 91 cm downstream of the orifice, 13 cm from the wall, and 25 cm from the fishway floor (Figures 3 & 5).

At weir 67, one Photosea Nighthawk SIT camera, rated at 0.01 lux with a field of view of 110° diagonal, 98° horizonal and 81° vertical, was suspended directly over the overflow on the south side of the weir. The cameras were 0.95 cm in diameter and 2.98 cm in length. The camera was first placed 129.5 cm above the weir and then lowered to 76.2 cm. During the fish ladder's last maintenance period, the top of the weir had been painted white (Figure 5).

At the vertical slot between pool 8 and 9, aluminum guides were installed in the winter of 1993-1994 to allow cameras and white panels with grid marks to be installed during the study. The white panels with grid marks were placed on the wall opposite the camera so that the fish had to swim between the white panel and the camera. This allowed fish location, velocities, and depths to be recorded. One camera was positioned downstream and one camera was positioned upstream of the vertical slot. These cameras were identical to the one used at the overflow weir. The cameras were 76.2 cm from the weir they faced and were 20.3 cm from the weir to which they were attached (Figure 6).

The equipment was washed with unscented soap to remove any human scent and then rinsed before being placed in the fishway. If the equipment was handled after being washed, rubber gloves were worn or the equipment was washed again. Except for installing the overflow weir camera, all adjustments were made after dark.

#### Information Recorded

The following data were collected from the video tapes: date, time, camera location, camera depth or station, fish species, the number of video frames the fish was in view, grid coordinates of the fish's nose (when possible) entering and exiting the camera's view to the nearest 7.6 cm, and description of what the fish was doing. For the submerged orifice, the grid coordinates of the fish entering and exiting the camera's view were not recorded. For the overflow weir, the time in the view (except for lamprey), the grid coordinates of the fish entering and exiting the camera view, and camera depth were not recorded.

Fish viewed on the tapes were identified into the following categories: teleosts, steelhead (*Oncorhynchus mykiss*), chinook salmon (*O. tshawytscha*), coho salmon (*O. kisutch*), sockeye salmon (*O. nerka*), unidentified salmonids, (*O. sp.*), black bass, (*Micropterus sp.*), Pacific lamprey (*Lampetra tridentatus*), and other fish. Other fish consisted mostly of northern squawfish (*Ptychocheilus oregonensis*), mountain whitefish (*Prosopium williamsoni*), peamouth (*Mylocheilus caurinus*), chiselmouth (*Acrochelilus alutuceus*), suckers (*Catostomus sp*), American shad (*Alosa sapidissima*) and fish that could not be identified.

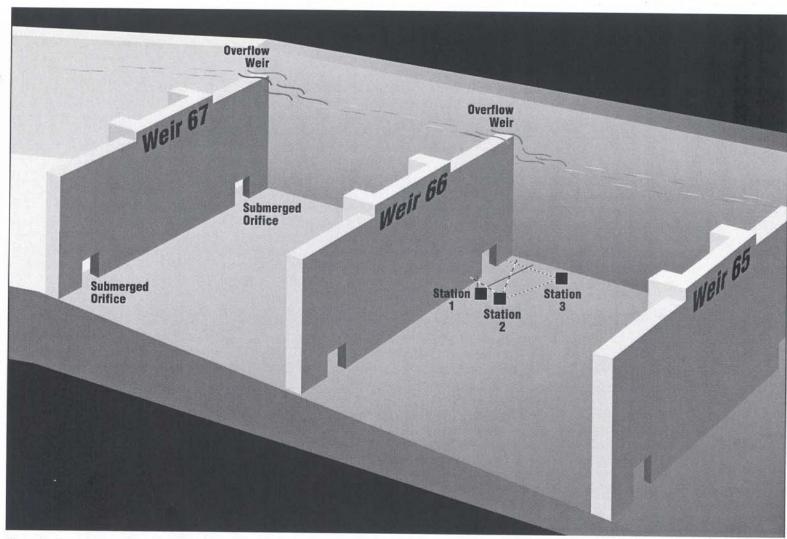


Figure 3. General view of the pool and weir type ladder showing the submerged orifices and overflow weirs in the Washington Shore ladder at the Bonneville Dam second powerhouse.

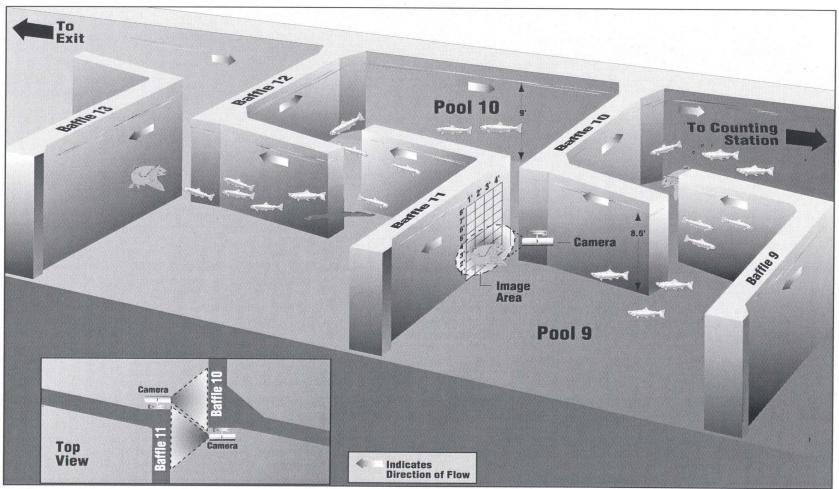
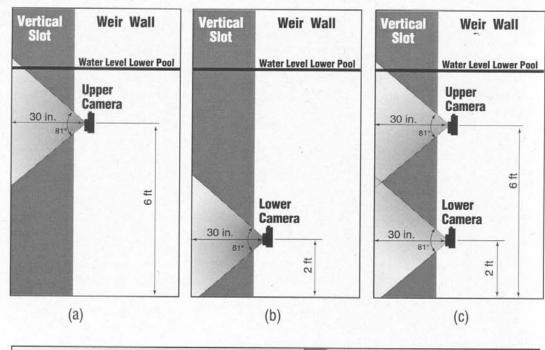


Figure 4. General view of the ladder showing locations of the cameras in the vertical slot at the Washington shore ladder at the Bonneville Dam second powerhouse.

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Figure 5. Slide and top view of the cameras that observed the submerged orifice and the overflow weir in the Washington shore ladder at the Bonneville Dam second power-house.



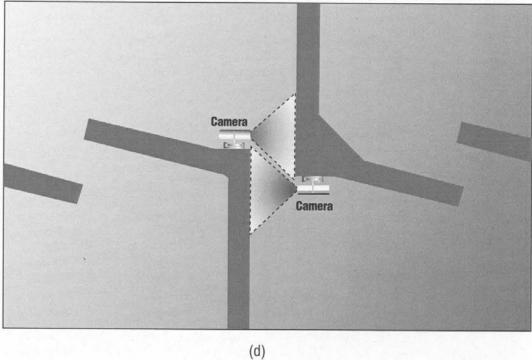


Figure 6. Side and top view of the cameras that observed the vertical slot at the Washington shore ladder at the Bonneville Dam second powerhouse.

The fish movements were categorized as follows:

- 1. up movement fish passing upstream through the area of interest
- 2. <u>down movement (tail first)</u> fish passing downstream through the area of interest with tail downstream
- 3. <u>down movement (head first)</u> fish passing downstream through the area of interest with head downstream
- 4. <u>no passage</u> fish comes into view of the camera swims around and then goes out of camera view without passing through the area of interest
- 5. <u>unknown</u> fish that entered the camera view but could not be placed into any of the above categories

# **Sampling Times**

At the submerged orifices, cameras were placed on 4 October 1993 in the ladder between weir 65 and weir 66 at camera station 1. On 7 October 1993, the cameras were moved to station 2. On 13 October 1993, the cameras were moved to station 3. On 19 October 1993, the cameras were removed. Video recording occurred from 5 to 17 October 1993 between 0400 h and 2000 h. Data were collected from all video recordings. The video recording was set on 24 hour recording mode for the first day and 6 hr recording mode for the remainder of the sampling.

At the overflow weir, the camera was placed on 12 July and lowered on 27 July. Video recording occurred on 12, 13, 15, 20, 27, 28 July, 14 August, and 15 September 1994. Data were collected from video recordings made on 12 July 1994 between 10:10 h and 16:20 h and on 15 July 1994 between 11:46 h and 17:55 h.

At the vertical slot, the cameras were placed the night before and moved the night after video recording. Video recording occurred on 15, 16, 29, 30 June; 12, 13, 27, 28 July; 24, 25 August; and 14, 15 September 1994 between 0400 h and 2000 h. We recorded for two consecutive days. The first day, cameras were placed 61 cm from the floor (Figure 6b) and the next day they were placed 182.9 cm from the floor (Figure 6a). The position of the camera (182.9 cm or 61 cm off floor) was alternated at the start of each consecutive pair of days. On 24 August 1994, a guide for the backdrop downstream of the slot broke off and was removed the next night. Therefore, the video recording on 14 and 15 September was taken with both cameras in the upstream position, one at 182.9 cm and the other 61 cm above the fishway floor (Figure 6c).

Data were collected from the first 8 hours of the 15 June video recordings for the areas both upstream and downstream of the vertical slot. After the 1120 h on the 15 June recordings, the only information recorded for shad was the number going upstream. On the 16 June tape, data were collected until 0830 h upstream and downstream of the slot. Between 0830 and 1120 h on 16 June, no data were collected. Between the 1120 h and the 1800 h on the 16 June tape, the only information recorded for shad was the number going upstream. On the 24 August and 14, 15 September tapes, data were collected from four to five randomly selected 15 min periods per day for

each camera.

#### Data Analysis

Fish velocity (cm/sec) through the vertical slot was determined by the time (number of video frames converted to seconds) that a fish took to travel between grid coordinates on the backdrop. The field of view near the camera is narrow, but farther away from the camera, the field of view becomes large. This caused fish to appear on the grid coordinates differently than if viewed on axis of the camera, a condition know as parallax. Parallax causes a fish swimming near the camera to appear to have traveled a distance up to three time greater then it actually did. Therefore, the actual fish velocity may be up to three times slower then reported.

The velocity through the submerged orifice was estimated at station 1 as the time from first appearance in the camera field until the fish passed the plane of the orifice (35.6 cm), and at stations 2 and 3 as the time from first appearance crossing the metal plate until it crossed the plane of the orifice (45.7 cm). At station 1, the edge of camera field of view was not parallel to the weir, so the farther away from the camera the fish entered the camera's viewing field, the greater the distance the fish was from the orifice.

To analyze their velocity and time spent in view at each location, the fish were separated into two categories, lamprey and teleosts.

The vertical distribution of fish entering the camera view for each side of the vertical slot was determined from 15 and 16 June 1994 tapes. Vertical distribution data were adjusted to equalize the sampling area of the camera and to equalize sample size. The adjustment was made because the cameras sampled at one depth one day and the another depth the next day. Data were adjusted to account for the differences in the number of fish present at the count station on these days (the number of fish at the count station was used to determine the number of fish present at the vertical slot). In addition, the camera's view is pyramidal in shape, small near the camera and large at the backdrop. To correct for the camera's view, the percentage of the slot viewed for each 7.6 cm depth was determined and then divided into the number of observed fish at that depth.

Water velocity in the vertical slot was measured with a Marsh McBirney model 201D portable water current meter on 27 September 1994. Velocity was measured 55 cm and 220 cm from the floor at 61 cm intervals horizontally through the area viewed by cameras.

#### **Results**

# **Submerged Orifice**

A total of 728 fish were observed in the two camera positions at the submerged orifices: 388 downstream of the orifice at weir 66 during 84 h and 6 min of video and 340 upstream of the orifice at weir 65 during 85 h and 4 min of video. Downstream of the orifice at weir 66, there were 8 Pacific lamprey, 44 unidentified salmonids, 107 steelhead, 6 coho salmon, 43 chinook salmon, 42 bass and 138 other fish. Upstream of the orifice at weir 65, there were 3 Pacific lamprey, 38 unidentified salmonids, 66 steelhead, 25 coho salmon, 70 chinook salmon, 36 bass and 102 other fish.

Eight Pacific lamprey were observed downstream of the submerged orifice at weir 66 for a net passage up through the orifice of 1. Of these lamprey, 4 were moving upstream, 3 were moving downstream head first, and 1 did not pass. The mean time in view was 50 s and the median time was 11.5 s. Three Pacific lamprey were seen upstream of the submerged orifice at weir 65 and none passed. The mean time in view was 267 s and the median time was 364 s (Table 1).

The 309 teleosts observed downstream of the submerged orifice at weir 66 stayed in view an average 1.1 s with a maximum time of 5.8 s. Of these teleosts, 52% passed upstream, 4% passed downstream head first, 3% passed downstream tail first, and 42% did not pass. Of the 134 teleosts for which velocities were calculated, the mean velocity was 86 cm/s with a maximum of 392 cm/s. The 337 teleosts observed upstream of the submerged orifice at weir 65 stayed a mean of 1 s in view. Of these teleost, 81% passed upstream, 2% passed downstream head first, 2% passed downstream tail first, and 15% did not pass. Of the 255 teleosts for which velocities were calculated, the mean velocity was 158 cm/s, with a maximum of 343 cm/s (Table 2).

#### Overflow Weir

Seventy fish were observed at the overflow weir 67 during 12 hours and 18 min, but only Pacific lamprey could be identified as to species. Of the 67 teleosts, 48% went upstream over the weir, 43% did not pass, and 9% went downstream. Three Pacific lamprey were seen, one went upstream, one went downstream and one did not pass. Time in view was not taken.

#### Vertical Slot

A total of 2,961 fish were observed at the vertical slot in two camera positions: 1,116 downstream of the slot during 26 h and 59 min of viewing and 1,845 upstream of the slot during 30 h and 57 min of viewing. Downstream of the slot, there were 758 American shad, 156 Pacific lamprey, 23 unidentified salmonids, 7 sockeye salmon, 34 steelhead, 2 coho salmon, 42 chinook salmon, and 94 other fish. Upstream of the slot, there were 517 American shad, 171 Pacific lamprey, 264 unidentified salmonids, 30 sockeye salmon, 144 steelhead, 39 coho salmon, 189 chinook salmon, and 491 other fish.

One-hundred-fifty-six Pacific lamprey were observed downstream of the vertical slot, but these had a

net passage upstream of 20. Of the 156 lamprey 43% passed upstream, 13% passed downstream head first, 17% passed downstream tail first, 8% passed with an unknown type of passage, and 19% did not pass. The mean time in view was 87.5 s and the median was 1.4 s. Of the 72 Pacific lamprey for which velocities were calculated, the mean was 187 cm/s with a maximum of 1,045 cm/s. Of these velocities, 80% had velocities below 274 cm/s and 90% below 396 cm/s. Of the lampreys going upstream, all had velocities less than 244 cm/c (Figure 7). As mentioned in the method section, actual velocities may have been three times slower then the calculated ones. One-hundred-seventy-one Pacific lamprey were observed upstream of the vertical slot for a net passage downstream of 35. Of the 171 lamprey, 27% passed upstream, 22% passed downstream head first, 26% passed downstream tail first, 5% passed with an unknown type of passage, and 20% did not pass. The mean time in view was 10.3 s and the median was 1.2 s. Of the 85 Pacific lamprey for which velocities were calculated, the mean was 159 cm/s with a maximum of 430 cm/s (Table 3, Figure 7). Of these velocities, 80% had velocities below 213 cm/s and 90% below 305 cm/s.

The 960 teleosts observed downstream of the vertical slot stayed a mean 1.4 s in view with a maximum of 8 s. Of these teleosts, 80% passed upstream, 1% passed downstream head first, 1% passed downstream tail first, 11% passed with an unknown type of passage, and 8% did not pass. Of the 737 teleosts for which velocities were calculated, the mean was 117 cm/s with a maximum of 914 cm/s. Of these velocities, 80% had velocities below 152 cm/s and 90% below 183 cm/s (Figure 8). The 1,674 teleosts observed upstream of the vertical slot orifice stayed a mean of 1.1 s in view. Of these teleosts, 73% passed upstream, 2% passed downstream tail first, 5% passed with an unknown type of passage, and 19% did not pass. Of 1,014 teleosts for which velocities were calculated, the mean was 230 cm/s with a maximum of 813 cm/s. Of these velocities, 80% had velocities below 305 cm/s and 90% below 335 cm/s (Table 4, Figure 8). The information by species is displayed in the Addendum Tables A1 - A7.

Of the teleosts on the downstream side of the vertical slot, 83% stayed in view less than 2 s, 95% less then 3 s, and 98.8% less then 4 s. Of these, 100% of salmonids stayed in view less then 2 s (Table 5). Of the teleosts on the upstream side of the vertical slot, 93% stayed in view less then 2 s and 98% stayed less the 3 s. Of these, 87% of salmonids stayed in view less then 2 s (Table 6).

Of the Pacific lamprey downstream of the vertical slot, 66% stayed in view less than 2 s, 86% stayed less than 4 s, 87% stayed less than 10 s, 91% stayed less than 70 s, and 96% stayed less than 200 s (Table 5). Of the Pacific lamprey upstream of the vertical slot, 70% stayed in view less than 2 s, 83% stayed less than 4 s, 90% stayed less than 10 s, 95% stayed less than 70 s, and 99% stayed less than 200 s (Table 6).

Fifty-four percent of teleosts, 51% of Pacific lamprey, 66% of American shad, and 4% of salmonids entered the camera's view downstream of the vertical slot at a depth less than 122 cm (Table 7, Figure 9, 10, 11, and 12). Forty-six percent of teleosts, 46% of Pacific lamprey, 75% of American shad, and 6% of the salmonids upstream of the vertical slot entered the camera's

view at a depth less than 122 cm (Table 8, Figure 9, 10, 11, and 12).

Median

Current velocity was measured on 27 September 1994 between 2000 h and 2200 h. The forebay elevations ranged between 22.98 m and 23.07 m msl. Velocity reading upstream of the slot ranged from 140 to 152 cm/s. Velocity reading downstream of the slot ranged from 168 to 189 cm/s.

Table 1. Passage behavior of Pacific lamprey at the submerged orifices between weir 66 and weir 67 in Bonneville's Washington shore ladder. Behavior was classified as upstream (up), no passage, downstream head first (Dn-hf), and downstream tail first (Dn-tf) movements. All fish Up No passage Dn-hf Dn-tf Time in view downstream of orifice (s) 0 3 8 4 94.1 Mean 0.2 8.8 50.4 SD 63.0 4.9 62.5 0.4 3.5 0.2 Min Max 137 13 137 Median 119.5 0.2 10 11.5 Time in view upstream of orifice (s) 0 Ν 3 0 3 Mean 266.7 266.7 219.8 219.8 SD Min 15 15 Max 421 421

364

Table 2. Passage behavior of teleosts at the submerged orifices between weir 66 and weir 67 in Bonneville's Washington shore ladder. Behavior was classified as upstream (up), no passage, downstream head first (Dn-hf), and downstream tail first (Dn-tf).

1 0 ;	,,,		,	/	
	Up	No passage	Dn-hf	Dn-tf	All fish
Time in view downstream of orifice (s)					
N	160	129	12	8	309
Mean	1.2	1.0	0.6	1.4	1.1
SD	0.51	0.8	0.3	0.9	0.7
95% confidence interval	0.1	0.1			0.1
Min	0.4	0.2	0.1	0.4	0.1
Max	3.4	5.8	1.2	2.8	5.8
Fish velocity downstream of orifice (cr	n/s)				
N	116	4	7	7	134
Mean	78	71	208	106	86
SD	39	28	120	63	56
95% confidence interval	7				9
Min	6	30	30	8	6
Max	274	89	392	213	392
Time in view upstream of the orifice (s	)				
N	273	51	7	6	337
Mean	0.9	1.4	0.5	1.1	1.0
SD	0.4	1	0.2	0.8	0.5
95% confidence interval	0.0	0.3			0.1
Min	0.3	0.3	0.2	0.4	0.2
Max	2.5	4.4	0.8	2.6	4.4
Fish velocity upstream of orifice (cm/s)					
N	243	3	6	3	255
Mean	160	130	115	66	158
SD	51	79	22	43	52
95% confidence interval	6				6
Min	32	43	80	27	27
Max	343	196	137	113	343

Table 3. Passage behavior of Pacific lamprey at the vertical slots above the fish count station in Bonneville's Washington shore ladder. Behavior was classified as upstream (up), no passage, downstream head first (Dn-hf), downstream tail first (Dn-tf), and unknown. Up No passage Dn-hf Dn-tf Unknown Total Time in view downstream of vertical slots (s) 30 21 26 12 156 Mean 22.5 393.3 0.5 2.6 122.6 87.5 SD 59.7 1958.8 0.2 9.5 70.1 861.5 95% confidence interval 14.3 135.2 Min 0.7 0.4 0.3 0.3 0.5 0.3 Max 305 10757 1.0 49.0 245 10757 0.5 Median 2.1 1.5 0.6 1.41 1.38 Fish velocity downstream of vertical slot (cm/s) 72 48 12 11 0 86 281 389 187 Mean 400 SD 35 207 325 208 95% confidence interval 10 48 Min 36 281 111 104 36 239 914 281 1045 1045 Max 76 281 354 261 105 Median Time in view upstream of vertical slot (s) 35 38 43 8 171 46 40.7 0.9 4.7 3.0 10.3 Mean 1.7 3.2 0.7 35.6 SD 1.4 69.2 16.3 0.4 22.9 0.2 5.3 95% confidence interval 4.9 0.4 0.4 0.4 0.4 0.4 0.4 Min Max 6.8 315 5.0 107 9.0 315.0 1.2 3.4 0.7 1.4 1.7 1.2 Median Fish velocity upstream of vertical slot (cm/s) 32 27 0 85 26 Mean 155 215 109 159 102 SD 76 53 91 95% confidence interval 36 19 20 112 44 20 Min 430 385 305 430 Max 130 198 103 141 Median

Table 4. Passage behavior of teleosts at the vertical slots above the fish count station in Bonneville's Washington shore ladder. Behavior was classified as upstream (up), no passage, downstream head first (Dn-hf), downstream tail first (Dn-tf), and unknown.									
	Up	No passage	Dn-hf	Dn-tf	Unknown	Total			
Time in view downstream of vertical slot (s)									
N	766	72	6	12	104	960			
Mean	1.5	1.4	0.8	1.1	1.0	1.4			
SD	0.8	1.1	1.0	0.4	0.6	0.8			
95% confidence interval	0.1	0.3			0.1	0.1			
Min	0.2	0.3	0.3	0.5	0.3	0.2			
Max	5.7	8.0	2.8	1.7	3.7	8.0			
Fish velocity downstream of	f vertical s	lot (cm/s)	-	-	-	-			
N	673	16	3	9	36	737			
Mean	115	104	518	169	114	117			
SD	57	36	369	48	53	65			
95% confidence interval	4				17	5			
Min	5	36	183	100	34	5			
Max	457	177	914	249	261	914			
Time in view upstream of v	ertical slot	(s)		_					
N	1226	319	6	41	82	1674			
Mean	1.0	1.3	0.7	2.0	0.9	1.1			
SD	0.5	1.0	0.4	1.2	0.6	0.7			
95% confidence interval	0.0	0.1		0.4	0.1	0.0			
Min	0.1	0.1	0.5	0.2	0.1	0.1			
Max	7.48	6.5	1.6	7.5	2.8	7.5			
Fish velocity upstream of v	ertical slot	(cm/s)							
N	965	13	4	25	8	1014			
Mean	234	186	268	127	198	230			
SD	92	87	195	53	40	93			
95% confidence interval	6					6			
Min	31	75	126	56	114	31			
Max	813	366	549	322	239	813			

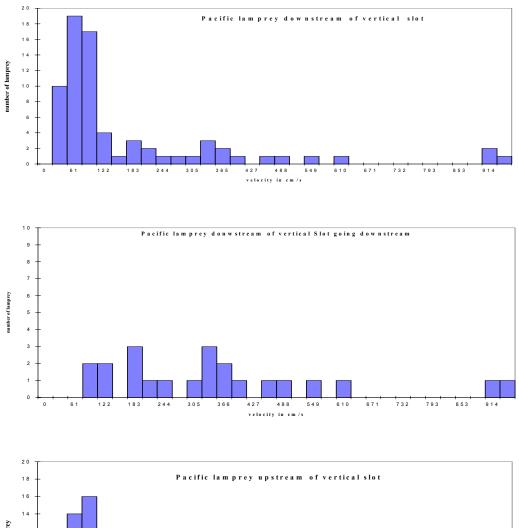
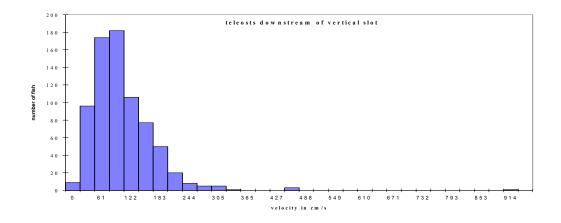


Figure 7. Frequency of velocity of Pacific lamprey downstream and upstream of vertical slot between pool 9 and 10 at Bonneville Dam second powerhouse Washington shore ladder in 1994.



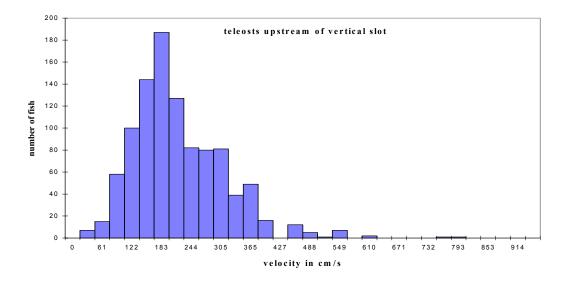


Figure 8. Frequency of teleosts velocities downstream and upstream of vertical slot between pool 9 and 10 at Bonneville Dam second powerhouse Washington shore ladder in 1994.

Table 5. Frequency of the time fish spent in the camera field downstream of the vertical slot in the Washington Shore ladder at Bonneville Dam second powerhouse. Pacific All Fish American Unidentified Sockeye Chinook Coho Other Time in s Steelhead Lamprey Salmonid Salmon Shad Salmon Salmon Fish 0.0-0.99 53 298 178 21 5 16 17 61 50 2 2 17 2 29 1.0-1.99 502 424 26 2.0-2.99 24 115 112 3 1 3.0-3.99 7 34 33 4.0-4.99 8 8 2 2 5.0-5.99 6.0-6.99 7.0-7.99 1 8.0-8.99 1 1 9.0-9.99 10.0-19.99 1 20-29.99 1 30-39.99 1 40-49.99 1 50-59.99 1 60-69.99 1 70-79.99 80-89.99 90-99.99 100-199.9 9 3 200-299.9 300-399.9 400-499.9 500-599.9 600-699.9 700-799.9 800-899.9 900-999.9

23

>1000

Total

156

960

758

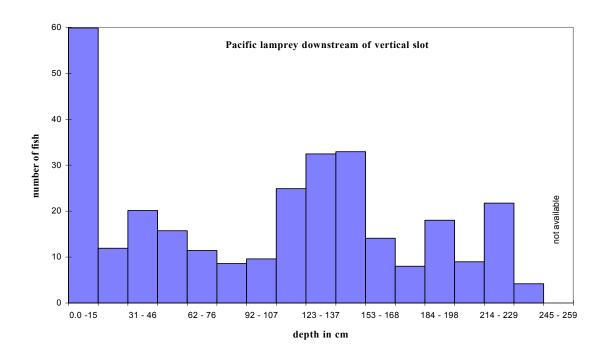
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34

2

Table 6. Frequency of the time fish spent in the camera field upstream of the vertical slot in the Washington Shore ladder at Bonneville Dam second powerhouse. Time in s Pacific All Fish American Unidentified Sockeye Chinook Steelhead Coho Other Salmonid Lamprey Shad Salmon Salmon Salmon Fish 0.0-0.99 73 847 272 108 14 41 31 5 375 1.0-1.99 46 711 238 123 13 126 94 19 96 2.0-2.99 14 87 22 2 20 13 9 15 6 3.0-3.99 9 3 17 7 3 4 4.0-4.99 4 7 2 3 5.0-5.99 3 2 1 4 1 6.0-6.99 1 7.0-7.99 2 1 1 8.0-8.99 9.0-9.99 1 10-19.99 4 2 20-29.99 30-39.99 40-49.99 50-59.99 60-69.99 1 70-79.99 1 80-80.99 90-99.99 1 100-199.9 6 200-299.9 300-399.9 400-499.9 500-599.9 600-699.9 700-799.9 800-899.9 900-999.9 >1000 Total 171 1674 517 261 30 189 144 39 491



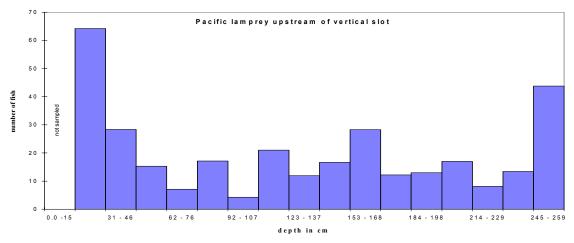
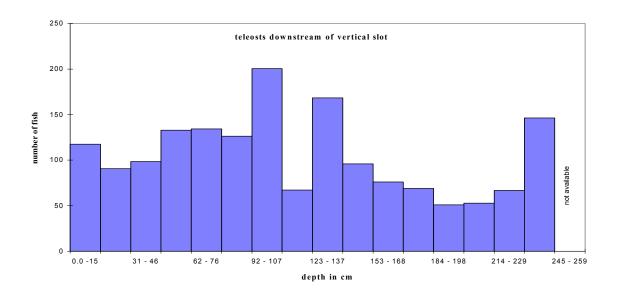


Figure 9. Frequency of depth of Pacific lamprey downstream and upstream of vertical slot between pool 9 and 10 at Bonneville Dam second powerhouse from 15 - 16 June 1994.



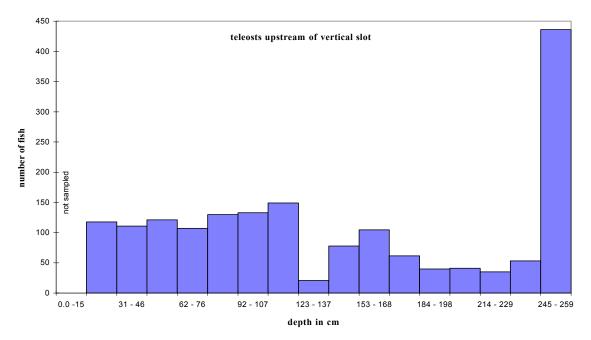
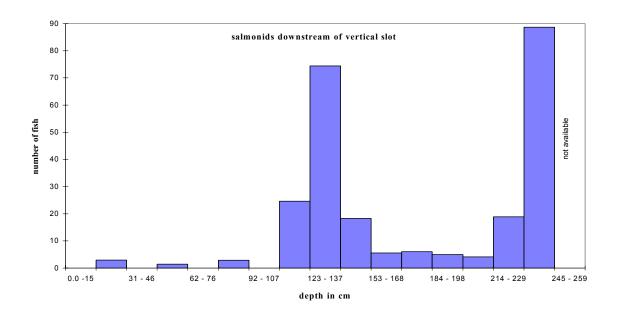


Figure 10. Frequency of depth of teleosts downstream and upstream of vertical slot between pool 9 and 10 at Bonneville Dam second powerhouse shore ladder from 15 - 16 June 1994.



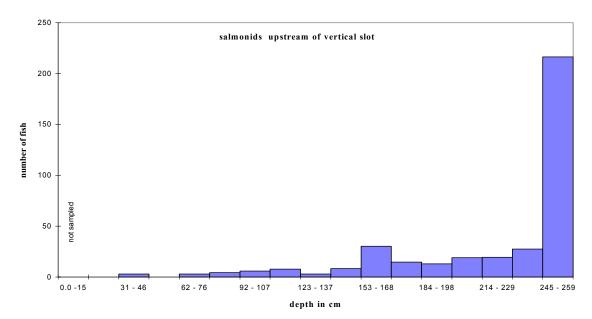
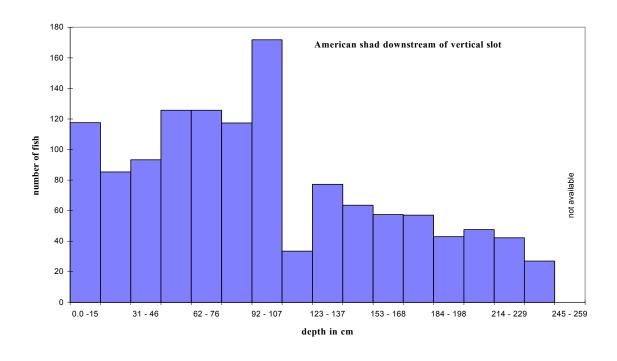


Figure 11. Frequency of depth of salmonids downstream and upstream of vertical slot between pool 9 and 10 at Bonneville Dam second powerhouse Washington shore ladder from 15 - 16 June 1994.



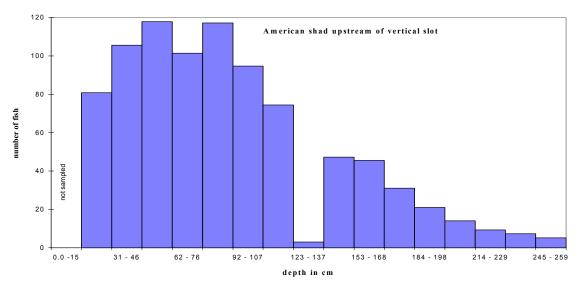


Figure 12. Frequency of depth of American shad downstream and upstream of the vertical slot between pool 9 and 10 at Bonneville Dam second powerhouse Washington shore ladder from 15 - 16 June 1994.

Table 7. Frequency of the depth of the fish entering the camera field downstream of the vertical slot in the Washington shore Bonneville Dam second powerhouse.

Depth in	Pacific	All Fish	American	Unidentified	Sockeye	Chinook	Steelhead	Coho	Other
cm	Lamprey		Shad	Salmonid	Salmon	Salmon		Salmon	Fish
0.0 - 15	56	87	87	0	0	0	0		0
16 - 30	16	87	84	3	0	0	0		0
31 - 46	20	102	96	0	0	0	0		6
47 - 61	14	115	109	1	0	0	0		4
62 - 76	9	139	134	0	0	0	0		6
77 - 91	13	123	110	0	0	1	1		14
92 - 107	7	158	132	0	0	0	0		25
108 - 122	14	115	96	0	0	0	3		22
123 - 137	32	168	77	14	11	24	25		17
138 - 152	28	117	58	8	2	11	13		25
153 - 168	22	78	66	0	1	2	0		9
169 - 183	6	78	62	0	0	3	5		8
184 - 198	16	57	46	0	2	1	4		4
199 - 213	13	58	53	0	0	0	2		3
214 - 229	15	63	44	1	0	10	5		3
230 - 244	11	77	33	7	0	18	6		12
245 - 259	0	84	0	39	0	11	6		22
Total	292	1706	1287	73	16	81	70	6	180

Table 8. Frequency of the depth of the fish entering the camera field upstream of the vertical slot in the Washington shore ladder ille Dam second powerhouse

Depth in cm	Pacific Lamprey	All Fish	American Shad	Unidentified Salmonid	Sockeye Salmon	Chinook Salmon	Steelhead	Coho Salmon	Other Fish
0.0 - 15									
16 - 30	56	40	16	0	0	0	0		24
31 - 46	32	143	127	0	3	0	0		13
47 - 61	14	111	107	0	0	0	0		4
62 - 76	10	115	112	1	0	0	0		2
77 - 91	14	124	113	3	0	1	0		7
92 - 107	10	143	116	0	3	0	3		21
108 - 122	16	149	76	3	2	2	0		65
123 - 137	16	64	28	0	3	2	0		31
138 - 152	17	78	47	3	6	0	0		22
153 - 168	25	100	46	8	11	2	5		27
169 - 183	18	68	27	3	9	5	4		19
184 - 198	12	449	29	3	3	3	2		8
199 - 213	15	42	18	0	3	9	3		9
214 - 229	12	36	11	1	1	11	9		3
230 - 244	12	45	7	4	0	11	9		13
245 - 259	16	107	7	8	0	39	19		35
260 - 274	31	353	3	62	6	50	42		189
Total	326	1767	890	99	50	135	96	0	492

#### **Discussion**

#### General

Past studies indicate fish take from 20 s to six min to pass through a pool in the ladder (Elling and Raymond 1956, Long 1959, Gauley 1960, Bell 1962, Weaver 1962, Thompson and Gauley 1963, and Monk et al. 1989). This study indicates fish were not spending much time in our sampling areas. Our sampling areas had high velocity flows which discouraged fish, except Pacific lamprey, from staying in the area. The mean times for the teleosts in view did not exceed 1.4 s at any location (Table 2 and 4). However, mean time in view for Pacific lamprey was as high as 267 s because some Pacific lamprey attached to the wall or floor for long periods of time.

Fish could have spent more cumulative time within the camera field than these results indicate. A fish was considered a different fish if it passed through the camera field and then reappeared. Observers believed that the same fish was occasionally observed several times.

It appears that, except for Pacific lamprey, submerged orifices and vertical slots would be good sites for PIT interrogator systems based on the time that a fish would spend in the electromagnetic field. Pacific lampreys could be discouraged from attaching near or in the PIT tag detectors. However, if the Pacific lamprey were not allowed to attach, their passage may be blocked. In addition, we could not determine from our data if the overflow weir was a good site because we had problems with the data.

The time in view is important because it provides an estimate of how long a fish may be exposed to the high electromagnetic field. The time in view is a maximum for a one time exposure because the PIT Tag systems should not extend 3 feet above or below the vertical slot as the camera field of view does.

The time in the PIT tag system may be calculated by knowing the fish's velocity and dimension of the PIT tag system. This time answers the question "do fish swimming through this area allow enough time to correctly interrogate a PIT tag?". The literature shows a maximum burst speed of 1,050 cm/s for the species we observed (Beamish, 1978) and we calculated a maximum velocity of 1,045 cm/s for lamprey going downstream. As we stated in the methods section, a fish's calculated velocity may be up to 3 times faster than its actual velocity because of parallax. Theoretically, the reported fish's velocity could be 3 times their actual velocity plus the water's velocity.

Because we were unable to correct for the error caused by parallax, we wondered if the velocities were reasonable and realistic. So, we compared velocities at the submerged orifice with those at the vertical slot which had similar current velocities. Mean velocities above and

below the vertical slot were 230 and 117 cm/s. Mean velocities above and below the submerged orifice were 158 and 86 cm/s. We feel these velocities are comparable but the velocity at the vertical slot may be exaggerated. Secondly, the distribution of velocities shows 95% are below 365 cm/s upstream of vertical slot and 95% were below 213 cm/s downstream of the slot. These velocities are within the upper limit of sustainable speeds of the fish stated in the literature which is an indication that they are reasonable. Many of the velocities calculated for the lamprey exceeded burst speeds for salmonids as cited in the literature. When velocities from lamprey going downstream are excluded, then the velocities do not exceed 244 cm/s (figure 9). These velocities still seem high but are more reasonable.

# Submerged Orifice

The mean times in view at the submerged orifices for teleosts did not exceed 1.1 s, and teleosts had mean velocities of 85 to 158 cm/s (Table 2). Thompson and Gauley (1965) show water velocities through the submerged orifice to be as high as 232 cm/s. Therefore, teleosts are not spending much time near the submerged orifice, and when they pass through the orifice, they do it quickly. Mean times in view for Pacific lamprey ranged from 56.4 to 267 s (Table 1); however only nine Pacific lamprey were observed. At station 2, more fish were seen traveling past the camera and turning into the middle of the ladder then at station 1. This would indicate that fish make a decision to pass through the orifice before they get within 30 cm of orifice.

#### Vertical Slot

Teleosts did not stay long in view at the vertical slot. The mean time in view did not exceed 1.4 s (Table 4) and mean velocities ranged from 116 to 232 cm/s. The maximum in either the upstream or downstream areas was 8.0 s. Water current measurements showed velocities as high as 189 cm/s. Therefore, teleosts pass through the vertical slot quickly. For information by species see the Addendum.

Most Pacific lamprey passed through the slot quickly, but a small percent stayed for extended times (Tables 5 and 6). Mean times for Pacific lamprey in view were as high as 87.5 s; median times were less then 1.4 s (Table 3). Although maximum time in view was as long as 3 hours, in both areas, 96% of the Pacific lamprey were in view less then 100 s, 90% were less then 60 s, and 83% were less then 3 s.

Many of the fish in the "unknown" movement category were observed by the lower downstream camera moving above the field of view. The upper downstream camera showed most fish passing upstream. This suggests that many fish in the unknown category were actually passing upstream. Upstream of the slot, fish were seen swimming towards the camera as they proceeded upstream. We assume these fish were going to the slow flow area on the inside of the pool.

The vertical distribution of the fish was affected by parallax, so only general observations may be made from the vertical distribution. The vertical distribution for the two day sampling period of Pacific lamprey and teleosts were generally uniform. Of the teleosts, most salmonids were seen at a greater depth than American shad which were shallower (Tables 7 and 8, Figure 9, 10, 11, and 12).

#### Overflow Weir

Fish at overflow weirs could not be identified to species because of the poor camera image. It was difficult at times to determine if what was being observed was a fish. The camera image may be improved by using polarizing filter, different cameras, different camera positions, and different viewing angles. The camera was placed over the last weir of the ladder because few air bubbles were present as water cascaded over the weir. Fish behavior upstream of this weir may be different than at most weirs, because fish are crowded to facilitate counting. Time in view was not taken because the fish would first appear near the downstream side of the weir. We feel that these fish could have been near the weir but not seen. If we had reported the time in view it may have been much shorter than what it actually is.

#### Miscellaneous

We found that the 6-hour recording mode was better for viewing fish than the 24-hour mode because it recorded rapidly swimming fish that the 24 hour mode missed. Recording during darkness failed to provide any useful information because of insufficient illumination.

We found that identification of fish could be difficult. Factors limiting identification are: seeing only parts of the fish, differing lighting conditions, and the changing profile of fish as they react to environmental conditions. A Washington Department of Fish and Wildlife fish counter, who was asked to identify fish we failed to identify, was able to identify more, but still was not able to identify all the fish. Further, we did not always agree on the species when the view was poor.

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# Addendum A.

Passage Behavior for Different Species of Fish

Passage behavior of unidentified salmonids at the vertical slots above the fish count station in Bonneville's Washington shore ladder. Behavior was classified as upstream (up), no passage, downstream head first (Dn-hf), downstream tail first (Dn-tf), and unknown. Table A1. Dn-tf Up Dn-hf Unknown Total No passage Time in view downstream of vertical slot (s) 2 0 0 23 Mean 0.7 0.5 1.2 0.7 SD 0.2 0.3 0.1 0.8 95% confidence interval 0.3 0.4 0.7 0.3 Min Max 1.0 0.6 1.7 1.7 Velocity of fish downstream of vertical slot (cm/s) 0 2 0 6 Ν 191 123 168 29 44 SD 33 95% confidence interval Min 157 100 100 229 229 146 Time in view upstream of vertical slot (s) 146 96 1 10 11 264 Mean 1.1 1.3 0.7 1.9 1.1 1.2 SD 0.5 0.8 1.2 0.6 0.7 95% confidence interval 0.2 0.1 0.3 0.2 0.2 0.2 0.2 Min 4.3 2.5 4.3 3.6 3.7 Velocity of fish upstream of vertical slot (cm/s) N 103 0 0 5 109 1 264 129 189 258 Mean SD 98 44 100 95% confidence interval 19 19 80 70 70 Min 549 549 166 Max

Bonneville'	s Washingtoi	nook salmon at th n shore ladder. E ad first (Dn-hf), o	Behavior wa	as classifie	d as upstream	(up), no
	Up	No passage	Dn-hf	Dn-tf	Unknown	Total
Time in view downstream of v	vertical slot (s	s)				
N	36	0	0	5	1	42
Mean	1.1			1.1	0.8	1.1
SD	0.3			0.4		0.3
95% confidence interval	0.1					0.1
Min	0.7			0.5		0.5
Max	1.6			1.5		1.6
Fish velocity downstream of v	ertical slot (c	em/s)		•	•	•
N	27	0	0	5	0	32
Mean	217			180		211
SD	83			54		79
95% confidence interval						27
Min	76			114		76
Max	457			249		457
Time in view upstream of vert	ical slot (s)	•			•	•
N	166	14	0	8	1	189
Mean	1.3	2.3		2.2	1.5	1.4
SD	0.4	1.7		0.6		0.7
95% confidence interval	0.1					0.1
Min	0.4	0.5		1.2		0.4
Max	2.6	6.5		2.9		6.5
Fish velocity upstream of vert	ical slot (cm/	s)	-	-	-	-
N	157	1	0	7	1	166
Mean	253	366		145	239	249
SD	74			84		77
95% confidence interval	12			1		12
Min	99			77		77
Max	499			323		499

Table A3. Passage behavior of steelhead at the vertical slots above the fish count station in Bonneville's Washington shore ladder. Behavior was classified as upstream (up), no passage, downstream head first (Dn-hf), downstream tail first (Dn-tf), and unknown.

	Up	No passage	Dn-hf	Dn-tf	Unknown	Total
Time in view downstream	of vertical	slot (s)				
N	30	1	0	2	1	34
Mean	1.1	0.8		1.1	0.9	1.0
SD	0.3			0.3		0.3
95% confidence interval	0.1					0.1
Min	0.6			0.9		0.6
Max	1.9			1.3		1.9
Fish velocity downstream of	of vertical s	slot (cm/s)				
N	20	0	0	2	0	22
Mean	168			186		169
SD	36			5		35
95% confidence interval						
Min	102			183		102
Max	229			189		229
Time in view upstream of v	vertical slot	: (s)				
N	86	40	1	8	9	144
Mean	1.3	1.6	1.6	2.2	1.5	1.5
SD	0.5	1.1		0.6	0.8	0.8
95% confidence interval	0.1	0.3				0.1
Min	0.6	0.2		1.2	0.6	0.2
Max	3.9	5.3		3.2	2.6	5.3
Fish velocity upstream of v	ertical slot	(cm/s)				
N	74	4	1	5	0	84
Mean	232	155	126	108		220
SD	98	93		26		100
95% confidence interval	22					21
Min	91	75		70		70
Max	813	289		144		813

Table A4. Passage behavior of sockeye salmon at the vertical slots above the fish count station in Bonneville's Washington shore ladder. Behavior was classified as upstream (up), no passage, downstream head first (Dn-hf), downstream tail first (Dn-tf), and unknown.

	Up	No passage	Dn-hf	Dn-tf	Unknown	Total
Time in view downstream	of vertical	slot (s)				
N	6				1	7
Mean	0.8				0.5	0.8
SD	0.2					0.2
95% confidence interval						
Min	0.7				0.5	0.5
Max	1.0				0.5	1.0
Fish velocity downstream of	of vertical	slot (cm/s)				
N	5				1	6
Mean	263				261	263
SD	110					99
95% confidence interval						
Min	189				261	189
Max	457				261	457
Time in view upstream of v	ertical slo	t (s)				
N	25	3	0	0	2	30
Mean	1.0	2.7			0.9	1.2
SD	0.5	1.2			0.4	0.8
95% confidence interval						0.3
Min	0.4	1.8			0.7	0.4
Max	2.4	4			1.2	4.0
Fish velocity upstream of v	ertical slot	(cm/s)				
N	24	0	0	0	0	24
Mean	288					288
SD	103					103
95% confidence interval						
Min	124					124
Max	499					499

Table A5. Passage behavior of coho salmon at the vertical slots above the fish count station in Bonneville's Washington shore ladder. Behavior was classified as upstream (up), no passage, downstream head first (Dn-hf), downstream tail first (Dn-tf), and unknown.

	Up	No passage	Dn-hf	Dn-tf	Unknown	Total	
Time in view downstream of vertical slot (s)							
N	2	0	0	0	0	2	
Mean	1.1					1.1	
SD	0.1					0.1	
95% confidence interval							
Min	1.0					1.0	
Max	1.2					1.2	
Velocity of fish downstream	n of verti	cal slot (cm/s)					
N	1					1	
Mean	157					157	
SD							
95% confidence interval							
Min							
Max							
Time in view upstream of v	ertical slo	ot (s)					
N	25	11	0	2	1	39	
Mean	1.4	2.9		3.2	2.8	2.0	
SD	0.8	1.0		0.3		1.1	
95% confidence interval						0.35	
Min	0.5	1.3		3.0		0.5	
Max	4.8	4.4		3.4		4.8	
Velocity of fish upstream of vertical slot (cm/s)							
N	24	1	0	1	0	26	
Mean	257	203		56		248	
SD	102					106	
95% confidence interval							
Min	110					56	
Max	610					610	

Table A6. Passage behavior of American shad at the vertical slots above the fish count station in Bonneville's Washington shore ladder. Behavior was classified as upstream (up), no passage, downstream head first (Dn-hf), downstream tail first (Dn-tf), and unknown.

	Up	No passage	Dn-hf	Dn-tf	Unknown	Total
Time in view downstream of	of vertical s	slot (s)				
N	612	61	3	3	79	758
Mean	1.6	1.5	1.1	1.0	1.0	1.5
SD	0.8	1.2	1.4	0.4	0.6	0.8
95% confidence interval	0.1	0.3			0.1	0.1
Min	0.3	0.3	0.3	0.6	0.3	0.3
Max	5.7	8.0	2.8	1.3	3.7	8.0
Fish velocity downstream of	f vertical s	lot (cm/s)				
N	573	15	2	0	30	620
Mean	104	105	686		102	106
SD	46	37	323		44	58
95% confidence interval	4				16	5
Min	5	36	457		34	5
Max	348	177	914		203	914
Time in view upstream of v	ertical slot	(s)				
N	498	5	0	2	12	517
Mean	1.0	1.1		1.8	0.9	1.0
SD	0.5	0.5		0.3	0.7	0.5
95% confidence interval	0.0					0.0
Min	0.1	0.5		1.6	0.1	0.1
Max	7.5	1.8		2.1	2.5	7.5
Fish velocity upstream of v	ertical slot	(cm/s)				
N	469	1	0	1	3	474
Mean	214	134		99	182	213
SD	80				58	80
95% confidence interval	7					7
Min	31				114	31
Max	549				215	549

Table A7. Passage behavior of other fish at the vertical slots above the fish count station in Bonneville's Washington shore ladder. Behavior was classified as upstream (up), no passage, downstream head first (Dn-hf), downstream tail first (Dn-tf), and unknown.

	Up	No passage	Dn-hf	Dn-tf	Unknown	Total	
Time in view downstream of vertical slot (s)							
N	61	8	3	0	22	94	
Mean	1.0	1.0	0.5		0.8	0.9	
SD	0.5	0.6	0.2		0.4	0.5	
95% confidence interval	0.1					0.1	
Min	0.2	0.5	0.4		0.3	0.2	
Max	3.2	2.2	0.7		2.1	3.2	
Fish velocity downstream of	of vertical s	lot (cm/s)					
N	43	1	1	0	5	50	
Mean	148	87	183		152	148	
SD	60				44	58	
95% confidence interval	18					16	
Min	39	87	183		102	39	
Max	274	87	183		215	274	
Time in view upstream of v	ertical slot	(s)					
N	280	150	4	11	46	491	
Mean	0.7	0.9	0.5	1.7	0.7	0.8	
SD	0.4	0.7	0.0	2.0	0.5	0.6	
95% confidence interval	0.1				0.4		
Min	0.1	0.1	0.5	0.4	0.2	0.1	
Max	3.7	3.8	0.5	7.5	2.5	7.5	
Fish velocity upstream of v	ertical slot	(cm/s)					
N	114	6	3	6	3	131	
Mean	247	182	315	137	203	240	
SD	125	72	209	31	32	124	
95% confidence interval	23	63				21	
Min	54	76	146	87	177	54	
Max	784	274	549	177	239	784	

# A STUDY TO DETERMINE THE BIOLOGICAL FEASIBILITY OF A NEW FISH TAGGING SYSTEM, PART III:

## **Development and Evaluation of PIT-tag Technology**

## PROGRESS REPORT 1994-1996

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#### **EXECUTIVE SUMMARY**

A multiyear program to evaluate the technical and biological feasibility of a new identification system for salmonids was established between the Bonneville Power Administration (BPA) and the National Marine Fisheries Service (NMFS) in 1983. This identification system is based upon a miniaturized Passive-Integrated-Transponder (PIT) tag. This report contains the results from 14 studies completed during 1994-1996 according to program requirements. These studies were divided into two groups, with nine described within the section titled "Development and Evaluation of PIT-tag Systems" and five described within the section titled "Activities at Columbia River Basin Dams."

Four major hydroelectric dams within the Columbia River Basin (CRB) have juvenile fish bypass/collection facilities that contain both PIT-tag interrogation and fish separation (diversion) systems. Interrogation systems energize PIT tags and process their identification codes into a usable form. Separation systems use fish diversion gates to separate PIT-tagged juvenile salmon from non-PIT-tagged fish and to separate targeted PIT-tagged fish from untargeted tagged and untagged fish. At the center of both interrogation and separation systems are dual-coil PIT-tag interrogation units. These units are described in this report.

## **Development and Evaluation of PIT-tag Systems**

The following nine research and development activities are summarized in this section: Underwater PIT-tag Interrogation Systems, Separation-by-Code System: Computer Program (BYCODE), Separation-by-Code System: Diversion Gates, Separation-by-Code System: An Evaluation Tool, Evaluation of Three Generations of 400-kHz Transponders, Evaluation of Generation-3B PIT Tags, Toxicity Evaluation of the Dye used to Detect Broken Tag Casings, Electromagnetic Field Effects on Reproducing Fish: Medaka (*Oryzias latipes*), and PIT-tag Retention in Adult Salmon. Essential elements and key results of each study are summarized individually below. Details on specific topics are presented in the corresponding reports for each study.

## **Underwater PIT-tag Interrogation Systems**

To minimize the stress of sampling fish with nets and trawls, NMFS and University of Washington staff designed and fabricated a prototype 400-kHz PIT-tag interrogation system that was attached to an open cod-end of a trawl net. This approach permitted fish to pass through the capture system unharmed and still allowed researchers to collect data on the migrating salmon. The system developed was towed by two boats to maintain net position and was evaluated on the Columbia River between 15 May and 27 June 1995 (total tow time was 72.6 hours). Information was obtained on 185 PIT-tagged fish.

A number of technical problems arose during the evaluation, but these should be correctable by modifying the net design and using a better sealant to prevent leaks in the interrogation housings. Another problem was that fish tended to congregate in front of the PIT-tag housings and were reluctant to swim through them. In post-evaluation tests, NMFS staff found that a 46-cm-diameter by 30-cm-long tunnel constructed of translucent material improved fish passage.

Despite the problems, the concept of using an open-ended net with an attached PIT-tag interrogation unit was shown to be feasible for the collection of data. When this system becomes operational with the recommended refinements, the information collected will significantly increase our knowledge of fish migrational patterns and behavior in the forebays of dams, in rivers, and estuaries. In addition, the electronic package, with minor modifications, could be attached to the cod-end of a fyke net or to a fish trap.

## **Separation-by-Code System: Computer Program (BYCODE)**

Separation-by-Code systems combine a computer program with one or more fish diversion gates. In 1994, NMFS issued a contract to Pacific Northwest National Laboratory (PNNL) to restructure the computer program so that tag databases could be larger, it would be easier to add new functions in the future, and the program could be more user friendly.

During 1994-1995, the following features were added to the computer program: 1) the maximum number of tag codes that could be stored in the tag database file was increased from 100,000 tags to over a million; 2) the ability to control two-way and three-way rotational diversion gates; 3) the ability to simultaneously control multiple fish diversion gates; 4) the ability to attach individual gate settings (i.e., delay and open times) to each coil above the different fish diversion gates; and 5) the ability to manually trigger the fish diversion gates from the keyboard.

Evaluating the Separation-by-Code system at Lower Granite Dam in 1995 was useful in revealing how the computer program needed to be modified to add the necessary flexibility to make it possible for multiple researchers to use the system simultaneously. These changes will be completed in 1996.

#### **Separation-by-Code System: Diversion Gates**

In 1994, NMFS started to address the need to route fish in multiple directions and to construct fish diversion gates for pipes. NMFS developed two-way and three-way rotational gates and side-to-side gates. General descriptions of the two types of diversion gates and how they operate are presented in the report. Evaluations showed that the side-to-side design has several advantages over the rotational design: it can be operated with the pipe at any degree of fullness, it causes less elevation loss, its fabrication is less costly because it requires fewer custom parts, and it is more easily maintained.

## **Separation-by-Code System: An Evaluation Tool**

Once the basic Separation-by-Code System was working, NMFS recognized that the computer program and test facility located at NMFS Manchester Research Station could be used to evaluate modifications being considered for installation at PIT-tag facilities in the CRB. To determine what modifications would be acceptable, the following comparisons were evaluated during 1994: 1) performance of single-read firmware versus double-read firmware at a water velocity of 4 m/second; 2) reading and separation efficiencies based on two versus four coils; 3) separation efficiencies at water velocities of 3 versus 4 m/second; and 4) separation efficiencies for two distances between last coil and diversion gate.

Tests were conducted with PIT-tagged sticks and coho salmon diverted by a slide gate. Reading efficiency (*RE*) was calculated by determining the percentage of tagged sticks or tagged fish read by at least one coil out of all possible PIT tags used in that trial. Separation efficiency (*SE*) for each trial was calculated using the theoretical and actual distributions of tagged sticks or fish within the two terminal holding areas based on which tags had been read. Thus, *SE* represented the percentage of correct actions for each trial.

Results for stick and fish trials using the four-coil arrangement at 4 m/second demonstrated that the *RE* and *SE* performance for double-read firmware was equivalent to that of single-read firmware. In the stick trials for both firmwares, all sticks were read and only one stick was not diverted successfully. Although more fish than sticks were missed, there were still no significant differences in *RE*s or *SE*s between single-read and double-read firmware. Furthermore, the double-read firmware did not produce a single erroneous tag code.

Thus, to avoid potentially harmful erroneous tag codes, NMFS supports incorporating double-read firmware into the interrogation systems at the CRB dams. However, after NMFS finished its tests, Destron-Fearing produced a new generation of 400-kHz tags that incorporated the more accurate cyclic-redundancy-check (CRC) method for error checking. They also wrote new firmware for these tags. Pacific States Marine Fisheries Commission (PSMFC) will install these CRC firmware chips into CRB PIT-tag interrogation equipment for the 1996 juvenile outmigration.

Increasing the number of interrogation coils from two to four coils significantly improved the ability to detect fish. At 3 m/second, average *RE* for the four-coil arrangement (98.3%) was significantly higher than average *RE* for the two-coil arrangement (93.6%). At 4 m/second, average *RE* for the four-coil arrangement (98.3%) was also significantly higher than average *RE* for the two-coil arrangement (93.8%). However, average *SE*s for fish were not significantly improved by utilizing all four coils at either 3 m/second or 4 m/second. The *SE*s for both the two- and four-coil arrangements ranged between 86.1 and 90.2%.

Although not statistically significant, average *SE*s were approximately 2% higher at 4 m/second than at 3 m/second whether the comparison was made for two or four coils. The higher water velocity created more turbulence within the rectangular flume, which appeared to cause fish to swim more actively to correct for the turbulence. NMFS recommends exchanging the rectangular flume for a pipe or round-bottom flume to reduce the turbulent water conditions to help improve separation at 4 m/second.

The shorter distance between the lower interrogation unit and slide gate yielded slightly higher SEs than the longer distance between the upper interrogation unit and the gate, but the increase in SEs was not significant at 3 m/second or at 4 m/second. However, if only those tags that were targeted to be diverted are considered, one can calculate a diversion efficiency by combining the REs and SEs (DE = percentage of the tags read that were programmed to be diverted and were successfully diverted). Calculated DEs showed that programmed fish that were read were separated significantly better over the shorter distance at both 3 m/second and 4 m/second. At both velocities, DEs were < 90% for the upper interrogation unit and close to 97% for the lower interrogation unit. Therefore, NMFS recommends that diversion gates be installed around 1 m (maximally 2 m) from the last coil in future PIT-tag separation system installations. This would permit a higher percentage of PIT-tagged fish to be successfully diverted.

## **Evaluation of Three Generations of 400-kHz Transponders**

The original 400-kHz PIT tags contained Atmill computer chips. When Atmill computer chips became unavailable, Destron-Fearing converted to Eurocell chips for their production tags. These did not perform well during the 1995 season, so Destron-Fearing tried Hughes Microelectronics computer chips. To avoid the in-season problems experienced in 1995, BPA asked NMFS to evaluate the new tags before PSMFC bought them. We designated tags containing Atmill computer chips as Generation-1 PIT tags, those with Eurocell chips as Generation-2 PIT tags, and those with Hughes chips as Generation-3 PIT tags. Performance of all three generations of tags was compared using the test facility at the NMFS Manchester Research Station. The effects of tag orientation (using tags at 45E orientation to simulate marginal reading conditions) and different excitation levels were examined.

With tags in the optimal 0° orientation, the resulting number-of-coils-read/tag averages for each generation were not significantly different. In contrast, when tags were tested at the 45° orientation, no Generation-3 tags and only one Generation-2 tag were read by all four coils, while most of the Generation-1 tags were read by all four coils. The resulting number-of-coils-read/tag averages for each generation were significantly different. A Tukey test separated the Generation-1 average from those of the other two generations. Other study results proved that poor performance by Generation-2 and Generation-3 tags was not due to their being turned off by high excitation power levels.

These results suggest that under normal monitoring conditions, Generation-3 tags would not be an improvement over Generation-2 tags and therefore, should not be purchased by PSMFC.

## **Evaluation of Generation-3B PIT Tags**

In another attempt to match the performance of Generation-1 tags, Destron-Fearing changed the signal modulation in its Generation-3 tags. These tags, designated Generation-3B, were evaluated in February 1996. In all tests, Generation-3B tags performed as well as Generation-1 tags, and significantly better than Generation-2 and Generation-3A tags. Therefore, NMFS recommends that PSMFC buy Generation-3B tags. Unfortunately, these tags were not available for the 1996 spring tagging season, but they were for the summer and fall tagging seasons.

## Toxicity Evaluation of the Dye used to Detect Broken Tag Casings

PIT tags are subjected to a series of quality-control tests during their manufacture. In one of these tests to identify broken casings, the newly produced tags are placed in a container with a dye and pressurized at 413.7 kPa (60 psi) for 2 hours. During this treatment, the dye penetrates broken tags and makes them easy to identify. At one time in the 1980s, a red dye was used that NMFS subsequently determined was lethal to fish. Therefore, when Destron-Fearing switched to a new tag manufacturing plant that used a different dye, NMFS again evaluated whether the new dye was toxic to fish.

Test fish were divided into four groups: 1) those injected with regular PIT tags that had been soaked in ethanol, 2) those injected with dyed PIT tags, 3) those injected with 0.5 mL of dye, and 4) those fin-clipped that represented controls. During a 72-hour observation period, no mortalities occurred and fish behavior was normal. Based on these results, the dye (mint green dye #1732) does not appear to be lethal to juvenile coho salmon or cause abnormal behavior; therefore, NMFS concluded that it is an acceptable dye.

## Electromagnetic Field Effects on Reproducing Fish: Medaka (Oryzias latipes)

The fisheries community has requested that interrogation systems for adult salmon be developed. However, during initial research, NMFS biologists observed that some volitionally migrating adults remained within the interrogation units for several hours. The potential for long exposure of migrating adult salmon to strong electromagnetic fields (EMFs) within interrogation units caused concern because the weakest calculated field strength within a PIT-tag interrogation unit is substantially higher than levels permitted under 1982 American National Standards Institute standards.

Therefore in 1991, NMFS initiated studies to examine whether fish were affected by exposures of up to 24 hours to 400-kHz or 125-kHz fields. It was recognized that to accomplish detection of adult salmon, it would be necessary to switch to a tag operating at a lower frequency. In 1991, most manufacturers were producing 125-kHz tags, so this was the frequency tested.

An earlier NMFS study used medaka (*Oryzias latipes*) as a surrogate for salmon. In this earlier study, there were differences in larval mortality between the control (20.1%) and EMF-exposed groups (27.3-33.7%) among the first-generation medaka offspring. In addition, the control group had fewer deformed hatched larvae (3.0%) than the EMF-exposed groups (5.0-11.5%). Although large, these differences were not significant because statistical power was low, with only six replicates completed. However, the results did suggest that EMF exposure may affect the survival and performance of first-generation offspring from EMF-exposed fish.

Therefore, NMFS designed a second experiment that would permit enough replicates (10) to provide the necessary statistical power for determining whether trends like those listed above are significant or merely due to normal biological variation (the control treatment was duplicated to give a better indication of what the normal level of biological variation was for this species). The modified experimental design also expanded on the first study to test not only tag-energizing frequency, but also field strength and field orientation. This report covers this second medaka experiment.

There were no significant differences between control and EMF-exposed treatments in any category (e.g., egg production/female, fertilization rates, larval mortality rates, deformity rates, overall survival). Duplicating the control treatment was critical for this study as the high standard-deviation values associated with averages for the controls showed a large amount of natural biological variation in this species. At this time, the results suggest no negative effects from exposure to the tested tag-energizing frequencies, field strengths, or field orientations. Assuming that these results are directly transferable, the results do not limit the design possibilities for developing adult salmon PIT-tag interrogation systems as long as adults will not be exposed continuously for longer than 24 hours. Exposures longer than 24 hours might not be a problem, but the effects of longer exposures would need to be tested if a design resulted in salmon being consistently exposed for >24 hours. NMFS recommends that the fisheries community continue pursuing its goal of interrogating adult salmon in fish ladders.

## PIT-tag Retention in Adult Salmon

The PIT tag is a reliable tool for identification of juvenile and adult salmon. However, an earlier NMFS study showed that up to 40% of female salmon and 20% of male coho salmon tagged as juveniles lost their tags during sexual maturation. Loss of PIT tags during sexual maturation limits the usefulness of these tags in situations where identification of mature adult fish is required (e.g., broodstock programs).

The PIT tag used in the CRB is encapsulated in biologically inert glass, and therefore it is usually found loose in the peritoneal cavity. PIT-tag manufacturers have found that by coating a tag with parylene or by adding a Teflon tip to the tag, they were able to stop PIT tags from migrating within small mammals. Therefore, NMFS investigated whether these tags, as well as acid-etched regular PIT tags, would reduce tag movement and loss within fish. These tags were compared to unmodified or regular PIT tags for tissue response and tag loss. This study was designed to test whether tissue response or encapsulation of the tag would retard tag loss during sexual maturation. A group of fin-clipped, untagged fish were included as controls for comparing growth and mortality rates between tagged and untagged fish.

Unfortunately, most test fish were killed during the first summer by a synergistic combination of stresses (tagging, anesthesia, elevated water temperatures). Consequently, the experimental design was drastically changed so that dead fish collected could be used to examine tissue response. Four time periods were established to examine how tissue response changed over time (from June 1995 to November 1996). Only one subsample of mature fish was conducted before all remaining fish were eaten by river otters.

Growth and survival results were not significantly different among the five treatment groups at any time during the study. Using the dead fish collected through 31 July 1995, it was possible to determine that consistent tissue response occurred earlier in the Teflon-capped (11 days post-tagging) and parylene-coated (15 days post-tagging) than in the acid-etched or regular PIT-tagged (both 22 days post-tagging) groups. Furthermore, both parylene-coated and Teflon-capped groups had half as many fish as regular and etched groups showing no tissue response during this first time period.

In all four of the time periods, the most consistent trend was that the regular PIT-tag group had the highest number of fish with no tissue response and the Teflon-capped group had the highest number with some tissue response. However, we still do not know if this tissue response will translate into better tag retention during sexual maturation.

With the fisheries community requiring the development of interrogation systems for adult salmon, NMFS recommends that this experiment be repeated. However, there are a few fish culture changes that NMFS recommends if this experiment were to be repeated. We recommend that tagging be done in early spring before water temperatures begin to rise. We also recommend that weights be taken on only 10% of the study fish instead of 100% because it is necessary to anesthetize fish longer when weights are being taken than if one is only tagging and taking lengths. We also recommend that smaller tanks be used so that it is easier to find the dead fish and that study fish be double tagged with a batch tag so that one could at least identify the treatment group on fish that have lost their PIT tags.

#### **Activities at Columbia River Basin Dams**

The following five research and development studies are summarized in this section: Review of PIT-tag Systems, Installation of PIT-tag Systems, Measurement of Radio-Frequency Emissions, Performance of Fixed-Reference Tags, and Evaluation of the Separation-by-Code System at Lower Granite Dam. Essential elements and key results of each study are summarized individually under the corresponding headings below. Details on specific topics are presented in the reports for each study that follow this summary.

## **Review of PIT-tag Systems**

NMFS worked with U.S. Army Corps of Engineers (COE) and its contractors in reviewing engineering concept drawings for Ice Harbor, John Day, The Dalles, and Bonneville Dams. NMFS input is critical in determining the number, placement, and installation of PIT-tag equipment. In August 1994, NMFS personnel joined a team of biologists from several fisheries agencies and the COE in reviewing future PIT-tag interrogation and fish separation needs for Lower Granite, Little Goose, Lower Monumental, and McNary Dams. The team's recommendations were presented to BPA in late 1994 and are summarized in the report.

## **Installation of PIT-tag Systems**

During 1994, new bypass/collection facilities for juvenile salmon were completed at McNary and Lower Monumental Dams. In 1995, an experimental site at Lower Granite Dam (GRX) was established as a platform for evaluating the rotational gates and the computer program (BYCODE) that controls fish separation. The GRX site operated independently of the main Lower Granite Dam site (GRJ). A similar experimental site (GOX) was established at Little Goose Dam in 1996.

## **Measurements of Radio-Frequency Emissions**

Radio frequency (RF) emissions from PIT-tag equipment must comply with Federal Communications Commission and National Telecommunications and Information Administration regulations for low-power electronics equipment. Tests were conducted to verify that the interrogation units met these requirements at Little Goose, McNary, and Lower Monumental Dams in 1994.

At Little Goose Dam, new aluminum shields had been fabricated for units that had exceeded the limit for RF emissions in 1993. When these units were retested in 1994, they all complied with the regulations once some exciter boards were corrected. At McNary Dam, despite the facility being new, all but one PIT-tag interrogation unit exceeded the limit for RF emissions. We found that the shields at McNary Dam lacked

welded seams as required by NMFS design specifications. The problem was corrected by retrofitting the shields to meet NMFS specifications. At Lower Monumental Dam, where the shields had been fabricated using NMFS specifications, all measurements of RF emissions were below the limit. Therefore, NMFS recommends that all future installations of PIT-tag systems include shields that meet NMFS design specifications.

## **Performance of Fixed-Reference Tags**

Fixed-reference tags test the operational status of each excitation/detection coil by simulating the passage of two PIT tags through that particular coil. During 1994, fixed reference tags were installed at five CRB dams. In 1995, NMFS requested that Destron-Fearing modify the tag codes of the fixed-reference tags so that all started with a common four-letter code. The change enabled fixed-reference tag codes to be easily identified from normal PIT-tag codes in the computer file. This change helped to improve on-site system analysis. The fixed-reference tag has become a critical maintenance tool for PSMFC.

## Evaluation of the Separation-by-Code System at Lower Granite Dam

To start transfer of this technology from the research and development stage at NMFS to the operations and maintenance environment at PSMFC, it was necessary to evaluate the system at a dam. The Separation-by-Code system was evaluated for its ability to direct PIT-tagged fish into five distinct pathways, and the rotational gates were evaluated for mechanical performance. To determine how fish behavior and fish density affected gate efficiencies, tests were conducted in April (low fish density) and May (high fish density) using two salmonid species.

In the April test, separation efficiencies for chinook salmon ranged from 93-97% while most separation efficiencies for steelhead were below 80%. The computer program was modified before the second test to permit setting different delay and open times for each species at each gate. Opening the gate longer for steelhead increased separation efficiency for the river-assigned fish from 73.3% to 89.7%. Unfortunately, because the water velocity was only around 1-1.5 m/second at the three-way gate, compared to almost 3 m/second at the two-way gate, there was not a similar increase for the left- and right-assigned fish (i.e., efficiencies remained below 80%). Therefore, NMFS recommends water velocities of 3 to 4 m/second for Separation-by-Code systems.

In general, the prototype rotational gates performed satisfactorily. However, it was observed during May that the rotational speed of the gates had slowed down relative to the April tests. The gates had probably slowed down from debris collecting in their mechanisms

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#### INTRODUCTION

In 1983, the National Marine Fisheries Service (NMFS) began a cooperative research program with the Bonneville Power Administration (BPA) to develop and evaluate a miniature 400-kHz, implantable Passive-Integrated-Transponder (PIT) tag for use with salmonids. Over the years, this program has encompassed many activities: evaluating different PIT tags, developing tagging techniques, investigating host responses to being tagged, developing PIT-tag interrogation and separation systems for dams, and coordinating the development of a PIT-tag information system (PTAGIS) for the Columbia River Basin (CRB).

In the CRB today, most PIT tags are implanted in juvenile fish. PIT-tag interrogation systems, which are located within juvenile fish bypass/collection facilities at federal hydroelectric projects, passively and non-intrusively collect information about individual fish as they migrate down river (Fig. 1). From 1987 through 1996, over 1.5 million juvenile salmon were marked with PIT tags. Both tagged and untagged salmon are subjected to the 400-kHz electromagnetic field (EMF) that energizes PIT tags as they traverse interrogation units.

Each energized PIT tag transmits a return signal at 40-50 kHz that contains the tag identification code. This return signal is received and processed by components of the interrogation system (Fig. 2; Prentice et al. 1990a). Along with the tag code, the time, date, and location of individual fish are recorded permanently in the PTAGIS database.

Four dams within the CRB have juvenile fish bypass/collection facilities that contain both PIT-tag interrogation and fish separation (diversion) systems. The latter systems mechanically separate PIT-tagged fish from non-PIT-tagged fish. The PIT-tagged fish are directed either back to the river or into special holding areas. This separation is accomplished without handling the fish, and the time, date, and location of individual fish are recorded as the fish pass through subsequent interrogation units. If tagged juvenile fish are returned to the river (e.g., below Lower Granite Dam), they can be subsequently re-interrogated at other downstream PIT-tag interrogation systems (Fig. 1).

At the center of both interrogation and separation systems for juvenile salmonids are dual-coil PIT-tag interrogation units (Fig. 2). All dual-coil PIT-tag interrogation units are assembled with the following standard components: 1) an aluminum shield to control errant radio frequency (RF) emissions and to provide weather protection for electronic components, 2) two excitation/detection coils (also called antennas) wrapped around a non-metallic fish passageway, 3) a tuner for each coil within the shield box, 4) a dual power supply, 5) a dual exciter board, 6) a power filter, and 7) a controller housing the tag-reading firmware and supporting electronics (Prentice et al. 1990a).

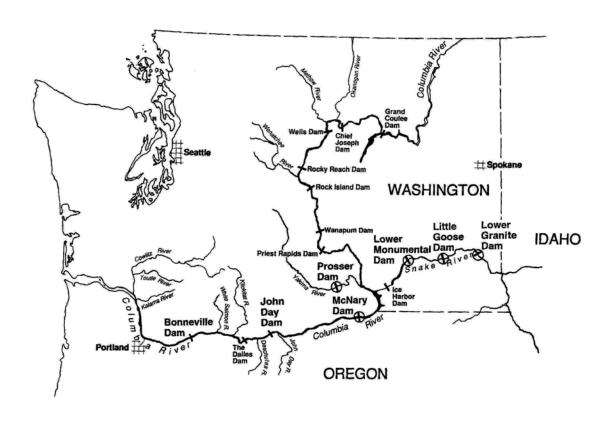


Figure 1. Hydroelectric dams in the Columbia River Basin. Those dams with PIT-tag interrogation systems are circled.

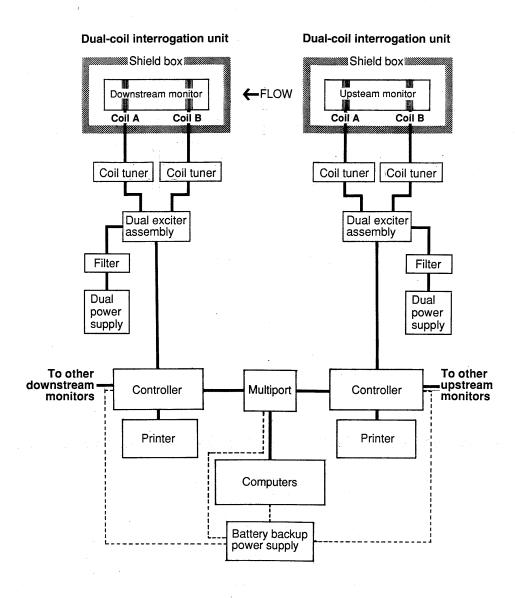


Figure 2. General schematic of a PIT-tag interrogation system like those used at Columbia River Basin dams.

To maximize the collection of data, the interrogation system is designed with redundant components to provide backup in case of component failure. For example, there are typically four coils (two dual-coil interrogation units) above a diversion gate so that if one set of two coils fails, information can still be collected and used to trigger the diversion gate.

This report covers a variety of work elements completed during 1994-1996. Other work elements completed during the same period were previously compiled into two separate reports because of their length and to expedite the transfer of information to the fisheries community. For convenience, this report is divided into two sections:

1) Development and Evaluation of PIT-tag Systems and 2) Activities at Columbia River Basin Dams.

#### DEVELOPMENT AND EVALUATION OF PIT-TAG SYSTEMS

## **Underwater PIT-tag Interrogation Systems**

#### Introduction

Various types of trawls, fyke nets, and traps are used to collect data on migrating juvenile and adult salmon in the CRB. Presently, all caught fish are handled to separate out the few fish of interest. During this sorting process, most fish, including unwanted bycatch, are severely stressed, and many are killed. To minimize the stress of the sampling process, NMFS and University of Washington staff designed and fabricated a prototype 400-kHz PIT-tag interrogation system that attached to an open cod-end of a trawl net. This approach would permit fish to pass through the capture system unharmed and still allow researchers to collect data on the migrating salmon.

#### **Methods and Materials**

The design and fabrication of the underwater PIT-tag interrogation unit was accomplished by personnel from NMFS Sand Point Electronics Shop. This segment of the study was supported by BPA. Adaptation of a Kodiak trawl net by adding extended wings to the main net was performed by University of Washington and NMFS staff. This group also evaluated the combined system (i.e., net and PIT-tag interrogation unit) on Lake Washington and on the Columbia River. This segment of the study was supported by the Portland District U.S. Army Corps of Engineers (COE).

The design for the underwater PIT-tag interrogation unit incorporated most of the standard electronic components that are used for interrogating juvenile salmon throughout the CRB (see Fig. 2). The computer, printer, power supply, exciters, and controller were maintained above water in an instrument barge that was towed behind the net. The coils and tuning circuitry were installed in waterproof housings that surrounded the fish passageways. Two of these housings (two antennas per housing) were placed side-by-side and then attached to the net in place of its cod-end section. The inside measurement of each fish passageway was 61-cm high by 20-cm wide by 89-cm long. The net and attached interrogation unit were towed by two boats (Fig. 3). Fish behavior in the net and near the PIT-tag interrogation unit was documented using video cameras, hydroacoustics, and divers. To determine if fish passing through the collection system were harmed, a sanctuary net was occasionally appended to the interrogation unit.

The combined system was evaluated on the Columbia River near Jones Beach (approximately 75 km from the mouth of river) between 15 May and 27 June 1995 (total tow time was 72.6 hours).

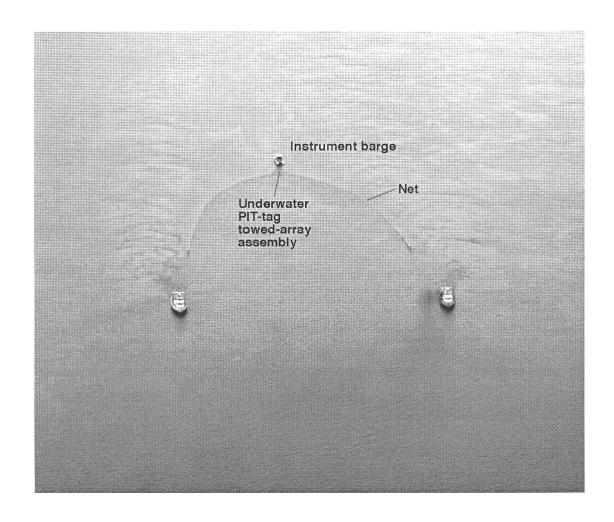


Figure 3. Photo of the underwater PIT-tag interrogation system in operation. Shown are the tow boats, net, and instrument barge. The PIT-tag interrogation unit is submerged and out of sight.

#### **Results and Discussion**

Information was obtained on 185 PIT-tagged fish. The information included their tag codes, the time and date of their interrogation, and the location where interrogation took place. In addition, 1,188 fish were captured in the sanctuary net for evaluation. Of the fish captured in the sanctuary net, 125 (10.5%) were descaled and 11 (0.9%) were injured. Over the entire study, 99 fish were killed; most were killed when the net wings were collapsed during retrieval.

A number of technical problems arose during the evaluation: 1) the large net size and the heavy interrogation housings made system deployment difficult; 2) it was difficult to maintain the net in its proper fishing configuration; 3) the interception of large debris at times was a problem; and 4) the interrogation housings leaked water, which caused electronic failures. To correct these technical problems, the net design is being modified and future interrogation housings will use a better sealant.

Another problem was that fish tended to congregate in front of the PIT-tag housings and were reluctant to swim through them. Most likely this behavior was due to both the small size of the tunnels, which was dictated by the short reading range of 400-kHz PIT tags, and the fact that the tunnels were constructed from non-translucent material. During August, NMFS staff tested several open cod-end designs to observe fish response and found that a 46-cm-diameter by 30-cm-long tunnel constructed of translucent material improved fish passage.

#### **Conclusions and Recommendations**

The concept of using an open-ended net with an attached PIT-tag interrogation unit was shown to be feasible for the collection of data. Compared to normal net sampling procedures, this approach will greatly reduced the impact on sampled fish. As indicated above, further refinements to the system are required before it can be considered ready for reliable field use. When this system becomes operational, the information collected will significantly increase our knowledge of fish migrational patterns and behavior in the forebays of dams, in rivers, and estuaries. In addition, the electronic package, with minor modifications, could be attached to the cod-end of a fyke net or to a fish trap.

Using additional electronics, future information on fish depth, environmental conditions, and sample locations could be obtained automatically. In addition, when the CRB converts from the present 400-kHz system to an ISO-based system operating at 134.2 kHz, the resulting longer read distance should enable further design changes to be made that will encourage fish to swim through the housings.

## **Separation-by-Code System: Computer Program (BYCODE)**

#### Introduction

A system that could divert specific PIT-tagged fish from other PIT-tagged or untagged fish would permit greater flexibility in addressing more specific questions in fish transportation, survival, and other studies. With this need in mind, NMFS developed and evaluated a prototype Separation-by-Code system during 1992-1993 (Prentice et al. 1994). Separation-by-Code systems combine a computer program with one or more fish diversion gates. The computer program uses the individual PIT-tag codes to separate desired or targeted PIT-tagged fish from untargeted tagged and untagged fish. When a particular fish is programmed to be diverted, the computer program sends an output signal to a gate controller that then sends the appropriate electrical signal to the fish diversion gate to make it open or rotate.

By the end of 1993, the computer program performed the basic data collection and separation functions, but was limited to Tag Database files of 100,000 codes and was difficult to use. Thus, in 1994, NMFS issued a contract to Pacific Northwest National Laboratory (PNNL) to restructure how the computer program was organized so that the Tag Database files could be larger, it would be easier to add new functions in the future, and the program would be more user friendly. For example, the Tag Database file was restructured so that it included the tag code of each targeted fish and an associated "Action code." An Action code was needed so that the program could quickly control the different diversion gates to get tagged fish to their appropriate destinations.

Action codes are decimal numbers (0-255) used to designate specific subsets of fish (e.g., different tagging sites, different treatments) that have the same set of actions applied to each tag within that subset throughout the entire facility. This way subgroups of fish can be treated differently through a dam (e.g., routed to different destinations). The actions (= output signals) for all Action codes for each coil within the interrogation system are defined in another section of the computer program. Output signals can also be defined for tag codes that are not in the Tag Database file. Internally, the computer program uses the Action code and not the individual PIT-tag codes to get PIT-tagged fish to their final destinations (e.g., whether an individual fish should exit to the river, to a barge, or to a particular sampling station). Below, the major modifications accomplished during 1994 and 1995 are discussed.

#### 1994

Most of 1994 was spent restructuring the computer program and adding a few critical features. The restructured program was given the name BYCODE (the program name is limited by DOS to 8 letters) as a shortened version of Separation-by-Code. Below are descriptions of some of the critical features added in 1994:

Increasing database storage from 100,000 tags to over one million--Being able to store a minimum of one million tag codes in the Tag Database file is necessary for the Separation-by-Code computer program to meet CRB needs. This number will enable tag codes from multiple years to be loaded into the Tag Database file at the same time. This feature will not only enable multiple investigators to conduct Separation-by-Code studies with juvenile fish at one site, but will make it possible to conduct a study with adult fish. The Tag Database file size was increased by incorporating a "bubble" sort approach. This approach meant that in a file with a million tags it would take a maximum of 12 comparisons to find the targeted tag code. The sort routine was evaluated for processing time using an oscilloscope. The bench test showed that the search time for tags, regardless of the number of tags in the database, did not exceed 1.5 milliseconds on a 486 PC computer. This speed should easily satisfy all Separation-by-Code applications in the CRB. No errors in the tag-code search and sorting process were detected in either laboratory or field testing of the program.

Adding control of two- and three-way rotational diversion gates--The test facility at the NMFS Manchester Research Station originally only had one slide gate, but it was expanded to permit testing of rotational diversion gates. The original computer program could only interface with slide gates and so computer code had to be written for the computer program to control the rotational gates.

Adding simultaneous control of multiple fish diversion gates--Since there were now multiple gates present at the test facility, we tested whether a programmable logic controller (PLC) would work as a centralized gate controller. A centralized gate controller would allow the computer to send different output signals to one location (the PLC) to open multiple gates simultaneously. More computer code had to be written to add the ability for BYCODE to interface with the PLC controller, but the PLC approach proved to be satisfactory and so PLCs were installed at dam sites starting in 1995.

Attaching individual gate settings (i.e., delay and open times) to coils--The fact that individual gate settings (i.e., delay and open times) could be assigned to each coil in interrogation units above each fish diversion gate meant that all four coils could be used to open a gate. In the current system installed at the dams, only the two lower coils open a slide gate, and the same gate settings are applied to both coils. By having the computer control the gate settings, different delay times could be set for each coil. If a tag code was successfully read at a second coil, the program deleted the gate-timing information for the first coil and inserted the new gate-timing information. That way the gate would be opened using the gate settings for the most downstream coil that read a fish. This is important because fish do swim in the flumes and if a diversion gate is opened too soon or too late, it could miss the targeted fish.

Adding a manual trigger for the fish diversion gates—In order to distinguish if a problem (e.g., diversion gate does not open) was due to the computer program or to diversion gate failure, the ability to manually trigger the fish diversion gates from the keyboard was added. This also enabled us to easily compare how the rotational diversion gates operated with different amounts of water flowing through them. This helped to improve the rotational gate designs.

**Improving the user friendliness of the program**--The previous program was difficult to use and so the program was designed to be menu driven to make it more user friendly.

## 1995

During 1994, the decision was made to test a complete Separation-by-Code system at Lower Granite Dam in 1995. A decision was also made that if everything went smoothly, the computer program would be installed at the main CRB PIT-tag sites in 1997. At the Lower Granite Dam Experimental site (GRX), two-way and a three-way rotational gates were installed as well as all of the electronic hardware and computers necessary for operating a site with 12 coils. Two fish tests were run. After the first, it became obvious that fish separation would be best if different gate settings could be applied to the two species being tested (steelhead and chinook salmon). Therefore, computer code was added for the ability to have multiple Diversion Units describing the same physical coils. These were referred to as logical Diversion Units. This helped improve the separation efficiency for steelhead.

A U.S. Fish and Wildlife Service (USFWS) researcher used the Separation-by-Code system at GRX after we finished our tests. He wanted to collect two different groups of fish, one of which had many more tag codes than the other. For this reason, he wanted to collect one in three fish from the large group and all fish from the smaller group. Initially, this was a problem because the program was written to apply the same ratio (1 in 3) to all gates and to all Action codes (or all of his test fish). A short-term solution for this research project was added, but we realized that the program needed to be changed to add the necessary flexibility to make it possible for multiple researchers to divert different ratios at the same and different diversion gates. The experience of using the program at a dam site also indicated several modifications that had to be completed before the computer program could be installed at the CRB dam sites as scheduled in 1997.

#### **Conclusions and Recommendations**

The restructuring of the program was helpful in adding more flexibility to the program and making it useful for fisheries researchers. When the results from the GRX evaluation and the USFWS study were presented at the PIT-tag workshop in January 1996, several researchers requested use of the computer program at two dams during 1996. To accommodate these requests, many of the identified modifications had to be immediately finished instead of waiting for the 1997 season. These modifications will first be tested at Manchester and then in the field using the researchers' studies. This will give us feedback from actual users and help us define how to improve the program so that it can satisfy their requirements. The development of this computer program is on schedule to meet the 1997 date for installation at the main Columbia River Basin sites.

## **Separation-by-Code System: Diversion Gates**

#### Introduction

As the Separation-by-Code system was developed, it became obvious that it would require PIT-tagged fish to be routed to new locations as they passed through the juvenile fish bypass/collection facilities. For example, fish could be routed to a fish holding tank so that researchers could examine their fish. However, in 1992-1993, only two types of fish diversion gates were available: a swing gate (Fig. 4) and a faster slide gate (Fig. 5). Both of the gates were designed for rectangular fish passage flumes and were limited to two-way fish diversion. Therefore in 1994, NMFS started to address the need to route fish in multiple directions and to construct fish diversion gates for pipes. NMFS developed rotational gates and side-to-side gates. Below is a general description of the two types of diversion gates.

#### Discussion

Two-way and three-way rotational gates were developed by NMFS between 1994 and 1996 (Fig. 6). Both rotational designs use an aluminum cylinder that has a portion cut away (about one third of the diameter). The cylinder is supported on both ends by a bearing assembly. Attached to one end of the cylinder is a sprocket that is in turn attached to a drive sprocket via a belt. The drive sprocket is controlled by a pneumatic piston that is operated with a motor. Upon receiving a signal from the computer, an electronic air valve opens and actuates the piston. A two-way piston is used for the three-way rotational gate and a one-way piston for the two-way gate. The two-way rotational gate is designed to rotate 180 degrees, while the three-way rotational gate rotates 160 degrees to the right or left of center. The mechanical movement of these diversion gates relies on pneumatic pistons that require 552-689 kilopascals (90-110 psi) of air pressure. The rotational design can be adapted to pipes of different diameters, water depths up to half a pipe depth, and for water velocities up to approximately 5 m/second.

The gates underwent initial mechanical, biological, and efficiency testing at Lower Granite Dam on the Snake River in 1995. A two-way rotational gate was also installed and evaluated at the experimental site at Little Goose Dam (GOX) in 1996 (the 1996-1997 Annual Report covers gate performance at the GOX site).

NMFS began development of two-way and three-way side-to-side gates in 1995 (Fig. 7). The general operating principal behind the system is that fish pass through a flexible hose section that is moved sideways to different passageways. The side-to-side design can be operated with the pipe at any degree of fullness at water velocities up to approximately 5 m/second. A two-way side-to-side gate was installed and evaluated at GOX for the 1996 season.

#### **Conclusions and Recommendations**

The side-to-side design has several advantages over the rotational design: it can be operated with the pipe at any degree of fullness, it causes less elevation loss, its fabrication is less costly because it requires fewer custom parts, and it is more easily maintained. However, the side-to-side design takes up more space and thus the characteristics of the particular installation site will dictate which design should be used. Technical and isometric drawings of these diversion gates are available through NMFS or BPA.

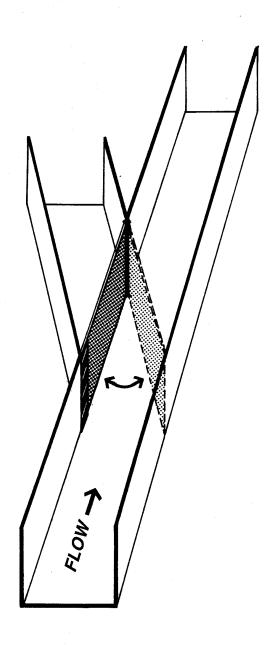


Figure 4. Diagram of a swing gate, a type of fish diversion gate.

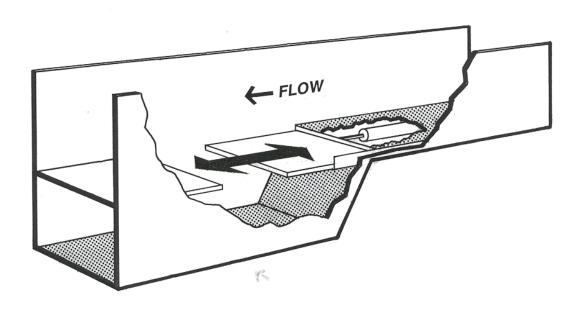


Figure 5. Diagram of a slide gate, a type of fish diversion gate.

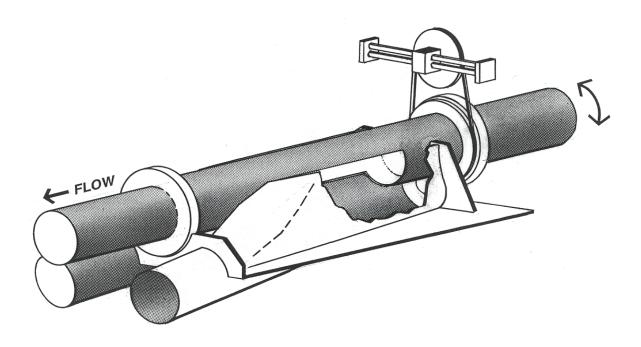


Figure 6. Diagram of a three-way rotational gate, a type of fish diversion gate.

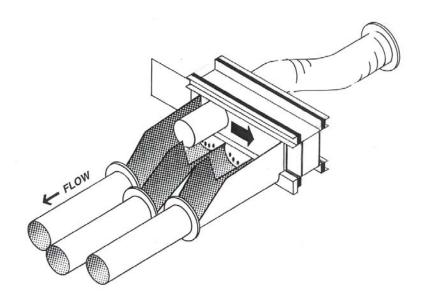


Figure 7. Diagram of a three-way side-to-side gate, a type of fish diversion gate.

## Separation-by-Code System: An Evaluation Tool

#### Introduction

To evaluate the Separation-by-Code system, NMFS constructed a PIT-tag test facility at the NMFS Manchester Research Station that included all of the standard components (e.g., interrogation units and a slide gate) installed in bypass/collection facilities at CRB Dams. During 1992-1993, this Manchester test facility was used primarily to evaluate the computer program BYCODE. Once the basic Separation-by-Code system was working, NMFS recognized that the computer program and the test facility could be used to evaluate modifications considered for installation at PIT-tag facilities. In fact in 1993, an adjustable slide gate and double-read firmware at water velocities of 3 m/second were evaluated with the system.

To determine what modifications would be acceptable for PIT-tag facilities, the following comparisons were evaluated during 1994: 1) performance of single-read firmware versus double-read firmware at a water velocity of 4 m/second; 2) reading and separation efficiencies based on two versus four coils; 3) separation efficiencies at water velocities of 3 versus 4 m/second; and 4) separation efficiencies for two distances between the last coil and diversion gate.

**Firmware-**Firmware located in computer chips on the reader cards inside of the 400-kHz tag reader is responsible for decoding the PIT-tag signals received from each coil and translating codes into a format usable by the PC computer. It is possible to insert different computer firmware chips. Single-read computer firmware chips (i.e., a chip that processes the first complete hexadecimal code received from a tag) are presently used in PIT-tag interrogation units at the dams. Single-read firmware processes each signal rapidly (12.5 milliseconds); however, single-read firmware also produces occasional erroneous tag codes (< 1% of all tag codes recorded). Although few erroneous codes are generated, there is a possibility that a particular erroneous code could be identical to a correct code, which would create a problem in a Separation-by-Code system.

To avoid erroneous tag-code readings, double-read firmware was written. Double-read firmware is slower (25-40 milliseconds), a factor that could be a problem under certain interrogation conditions, and thus it needs to be evaluated thoroughly before it can be installed at the CRB sites. Double-read firmware read PIT-tag codes as well as single-read firmware at 3 m/second (Prentice et al. 1994), but before it could be installed at the dams, it needed to be evaluated at 4 m/second, which is the fastest water velocity likely to be encountered within any bypass/collection facility in the CRB.

Reading and separation efficiencies based on two versus four coils—Reading efficiency (RE) was calculated by determining the percentage of tagged sticks or fish read by at least one coil out of all possible PIT tags used in that trial. When the test facility had two coils, the RE for fish was below the acceptable performance rate for the

CRB (≥ 95%; Prentice et al. 1994). Since at most dams there are four coils above each slide gate, NMFS installed a second dual-coil interrogation unit at the test facility in 1994. Four coils should increase the chances of reading a tagged fish when its orientation is satisfactory and permit more time for fish swimming side-by-side to disperse. We needed to confirm that a 4-coil arrangement, connected to the unique hardware of the Separation-by-Code system, would generate acceptable *RE* levels.

The installation of the second dual-coil interrogation unit also permitted testing whether higher separation efficiencies are yielded when the slide gate is triggered by all four coils instead of only two coils as is currently done at the dams. Separation efficiency (*SE*) for each trial was calculated using the theoretical and actual distributions of tagged sticks or fish within the two terminal holding areas based on which tags had been read.

Each PIT-tagged stick or fish that was programmed to be separated could follow one of four scenarios: 1) be read and be separated (correct action), 2) be read and not be separated (wrong action), 3) not be read and be separated (wrong action), and 4) not be read and not be separated (correct action). In scenario 4, the PIT-tagged stick or fish was acting as an untagged fish or as a PIT-tagged stick or fish that was not programmed to be separated. Therefore, fish or sticks in this scenario should not have been separated. Thus, *SE* represents the percentage of correct actions for each trial. Tags that were not read would lower *RE*, while *SE* was determined after incorporating the *RE* information.

**Separation efficiencies at water velocities of 3 versus 4 m/second--**Most of the 1992-1993 fish trials had been conducted to define procedures for running fish trials. They also yielded *RE* data, but only a few yielded *SE* data. Therefore, in 1994 we focused on running fish trials to learn how to achieve high *SE* values with the Separation-by-Code system. The earlier trials had revealed two reasons why fish often produce low *SE* values: fish exited in groups and they swam in the flume (Prentice et al. 1994).

Fish exiting in groups create a problem because if the gate opens for a targeted fish, some or all of its companions are also separated. However, this problem cannot be avoided with the current designs of fish/debris separators. Swimming in the flume can result in fish programmed to be diverted missing the slide gate and fish not programmed entering the slide gate. Fish were observed swimming in the flume at velocities of 3 m/second. Since most juvenile salmon cannot easily swim for long at velocities of 4 m/second, we investigated whether the higher water velocity might improve *SE*s for fish.

**Separation efficiencies for two distances between the last coil and diversion gate--** Prentice et al. (1994) also suggested that *SE*s for tagged fish might be improved if the distance between the last coil and slide gate was minimized. The installation of the second dual-coil interrogation unit made it possible to compare two different distances by triggering the gate with either the two upper coils or the two lower coils.

#### **Methods and Materials**

**Test facility-**The test facility, which simulates a portion of a bypass/collection facility, was modified in 1994 (Fig. 8). It was enlarged to evaluate prototype three-way fish diversion gates (e.g., the rotational gates). Large and small pipe sections were added for these evaluations. The large pipe section could be used for testing gates or coils measuring 25 or 30 cm in diameter and the small pipe section could be used for testing gates or coils measuring 10 or 15 cm in diameter. Furthermore, both pipe sections could be raised or lowered with pulleys to test different water velocities and hydraulic conditions. A third pump was installed to increase water flow during tests requiring the larger pipe section and 4-m/second water velocity.

Several changes were made to the original rectangular flume. A second dual-coil interrogation unit was installed whose final coil was 1.7 m above the slide gate compared to the 3.3-m distance of the original interrogation unit. A PLC was installed to replace an older-style slide-gate controller. This allowed all of the gates to be controlled with a centralized gate controller. Aluminum covers were built to be placed over the main slide-gate flume to darken the flume to the same level as the interrogation units during tests using fish.

**Evaluating the modifications**-The same general procedure was used for evaluating the four modifications to the PIT-tag system described above (i.e., firmware, number of coils, water velocity, distance between the last coil and diversion gate). Tests were conducted with PIT-tagged sticks and juvenile coho salmon (*Oncorhyncus kisutch*) whose fork lengths ranged from 150 to 225 mm. Tagged sticks were employed because both their rate of entry and orientation could be better controlled than with fish. Fish often passed through an interrogation unit in groups and at various angles; both of these can potentially reduce *RE*s and *SE*s. Therefore, modifications were first tested with sticks followed by tests with fish.

More stick trials than fish trials were conducted to evaluate the four modifications because of the time it took to perform fish trials (Table 1). Before each trial, the test facility was configured for that particular evaluation (i.e., depending on the trial, different coils would be turned on or off, different water velocities would be used, different reader firmware installed, etc). Each trial consisted of 50 tags in which 20, 50, or 80% of the PIT-tagged sticks or fish per trial had been programmed to be diverted.

Fish and stick tag-codes were appended to an existing Tag Database file containing 200,000 tag codes. Sticks or fish were then randomly introduced into the flume leading to the PIT-tag interrogation coils and slide gate. After passing through the slide-gate system, the final destinations of the individual sticks and fish were determined. This actual distribution was then compared to the theoretical distribution determined by the computer program for calculating *SE*s. Since it was necessary to increase the opening

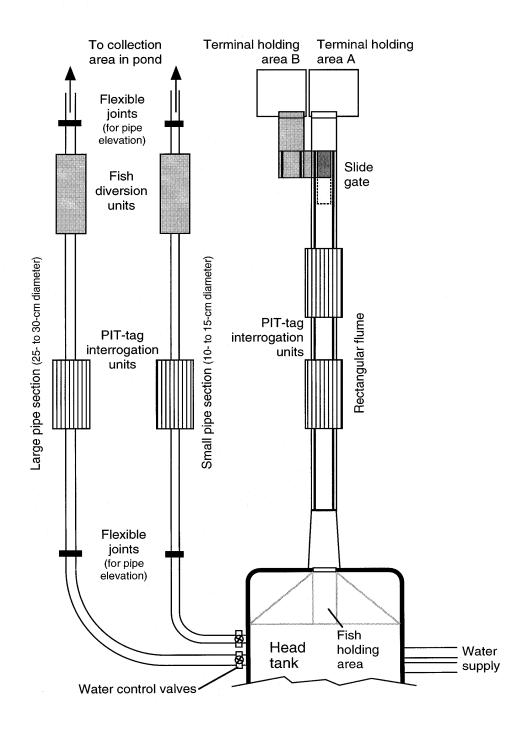


Figure 8. Diagram of the enlarged PIT-tag test facility located at the NMFS Manchester Research Station.

Table 1. The conditions and numbers for the different stick and fish trials performed for the four evaluations. Each trial used 50 PIT tags.

Possible configurations	Stick trials	Fish trials
Upper two-coils, SRa, 3 m/sec	15	5
Upper two-coils, DR <sup>b</sup> , 3 m/sec	15	5
Lower two-coils, SR, 3 m/sec	15	5
Lower two-coils, DR, 3 m/sec	15	5
Upper two-coils, SR, 4 m/sec	15	5
Upper two-coils, DR, 4 m/sec	15	5
Lower two-coils, SR, 4 m/sec	15	5
Lower two-coils, DR, 4 m/sec	15	5
Four-coils, SR, 3 m/sec	30	10
Four-coils, DR, 3 m/sec	30	10
Four-coils, SR, 4 m/sec	30	10
Four-coils, DR, 4 m/sec	30	10

a SR is an abbreviation for single-read firmware.
 b DR is an abbreviation for double-read firmware.

of the slide gate from 45 to 58 cm to accommodate the 4-m/second water velocity, this slide gate opening was used for all trials.

**Statistics**--There was no difference in the results whether 20, 50, or 80% specifically tagged sticks or fish were separated, and consequently the data from all of these trials were combined to evaluate the main elements. Independent t-tests were used to compare REs and SEs for 1) the two firmwares at 4 m/second; 2) two versus four coils; 3) 3- versus 4-m/second water velocities; and 4) using the upper or lower interrogation units to trigger the slide gate. Significance was established at  $P \le 0.05$ .

#### **Results and Discussion**

Modifications to the test facility proved to be satisfactory. The flexibility in the facility design allowed all of the reported evaluations to be conducted and provides a platform from which future tests can be conducted in both rectangular flumes and pipes of varying sizes. Below the four evaluations are presented separately.

**Firmware--**In all of the single-read computer firmware stick and fish trials, 0.3% (n = 27) of the tag codes were incorrectly processed. The erroneous tag codes typically contained single hexadecimal digits that have been misread and replaced. No erroneous tag codes were produced by the double-read firmware.

Results for stick and fish trials using the four-coil arrangement at 4 m/second demonstrated that the RE and SE performance for double-read firmware was equivalent to that of the single-read firmware (Table 2). In stick trials for both firmwares, all sticks were read, and only one stick was not diverted successfully. Although more fish than sticks were not read and missed by the slide gate, there were still no significant differences in REs (P = 0.883) or SEs (P = 0.561) between single-read and double-read firmware. For both types of firmware, average REs were approximately 98% (based on four coils) and average SEs were close to 88%.

It is not surprising that double-read firmware did well at 4 m/second because each PIT tag remains within a coil's electromagnetic field for almost 80 milliseconds at 4 m/second, and thus there is sufficient time for PIT-tag codes to be processed by double-read firmware, which takes a maximum of 40 milliseconds to process tag codes. To avoid potentially harmful erroneous tag codes, NMFS supports incorporating double-read firmware into the interrogation systems at the CRB dams.

However, after NMFS finished its tests, Destron-Fearing (the manufacturer of the PIT-tags used in the CRB) produced a new generation of 400-kHz tags. In these new Generation-2 PIT tags, Destron-Fearing replaced the Manchester encoding error-checking method with the faster and more accurate cyclic-redundancy-check (CRC) method. The CRC method will produce an almost errorless format (1 error in 10<sup>6</sup> reads). Destron-

Table 2. Overall average reading efficiencies (*RE*s) and separation efficiencies (*SE*s) for the two firmwares. Standard deviations are shown in parentheses. Probability values are derived from t-tests.

	Overall <i>RE</i> (%)	Overall SE (%)
Sticks		
Single-read firmware(4 m/sec, 4 coils)	100.0 (0.0)	99.9 (1.3)
Double-read firmware (4 m/sec, 4 coils)	100.0 (0.0)	100.0 (0.0)
Probability value	1.000	0.321
Fish		
Single-read firmware (4 m/sec, 4 coils)	98.2 (3.3)	88.9 (6.1)
Double-read firmware (4 m/sec, 4 coils)	98.4 (2.6)	87.3 (4.6)
Probability value	0.883	0.561

Fearing also wrote new single-read firmware for these Generation-2 tags that promises to eliminate the erroneous tag-code problem and read tags in 19 milliseconds. Pacific States Marine Fisheries Commission (PSMFC) will install these CRC firmware chips into CRB PIT-tag interrogation equipment for the 1996 juvenile outmigration.

Because there was no difference in performance, the single-read and double-read firmware results were combined for the other evaluations.

Reading and separation efficiencies based on two versus four coils--Although individual coils often had REs below 100%, the four-coil combination detected all but one tag that was introduced into the interrogation system during 120 stick trials (60 trials each at water velocities of 3 and 4 m/second) that represented a total of 6,000 tags (Table 3). When only two of the four coils were active, several tags were not read as the flowing water would change the tag orientation, especially at 4 m/second (97.9  $\pm$ 2.9%;  $\pm$ SD). Consequently, average RE for the four-coil arrangement was significantly higher than for the two-coil arrangement at 4 m/second (P < 0.001). Sticks were individually introduced at 1- to 2-second intervals, and therefore if they were read then the slide gate usually separated them successfully. There was a significant difference between the two coil arrangements at 3 m/second (P < 0.001); however, with the lower value being so high at 99.2%, there does not seem to be a reasonable explanation for the statistical difference.

Increasing the number of interrogation coils from two to four significantly improved the ability to detect fish (Table 3). At 3 m/second, average RE for the four-coil arrangement (98.3  $\pm$  4.5%) was significantly higher (P = 0.024) than average RE for the two-coil arrangement (93.6  $\pm$  7.3%; Table 3). At 4 m/second, average RE for the four-coil arrangement (98.3  $\pm$  2.9%) was also significantly higher (P < 0.001) than average RE for the two-coil arrangement (93.8  $\pm$  4.7%). However, average SEs for fish were not significantly improved by utilizing all four coils at either 3 m/second (P = 0.322) or 4 m/second (P = 0.171; Table 2). The SEs for both two- and four-coil arrangements ranged between 86.1 and 90.2%.

Using four coils instead of two coils did significantly increase the *RE* for fish. The four-coil arrangement increased *RE*s significantly because fish rarely travel side-by-side for long. However, *SE*s were not increased. Matthews et al. (1990) demonstrated that the number of fish separated each time a slide gate opens is basically a constant, which depends on the density of fish passing through the flume. The value of this constant, which will be directly proportional to the *SE* value, will be different for each gate setup (e.g., it will depend on such things as gate delay and open times, distance from last coil, and water velocity). In other words, *SE* values did not increase as more targeted tags were read with the four-coil arrangement because the same ratio of targeted and untargeted fish were separated each time the slide gate opened. Although the two-and four-coil arrangements yielded similar *SE*s, overall more targeted fish would be separated with the four-coil arrangement than a two-coil arrangement because more of them would be read.

Table 3. Overall average reading efficiencies (*RE*s) and separation efficiencies (*SE*s) for the 2-coil and 4-coil configurations. Standard deviations are shown in parentheses. Groups were statistically compared using t-tests.

	Overall <i>RE</i> (%)	Overall SE (%)
Sticks		
Two-coil arrangement (3 m/sec, upper or lower)	99.6 (2.8)	99.2 (1.3)
Four-coil arrangement (3 m/sec)	99.9+ (0.3)	99.9* (0.4)
Two-coil arrangement (4 m/sec, upper or lower)	97.9 (2.9)	100.0 (0.0)
Four-coil arrangement (4 m/sec)	99.9* (0.5)	99.9+ (0.3)
Fish		
Two-coil arrangement (3m/sec, upper or lower)	93.6 (7.3)	87.9 (5.2)
Four-coil arrangement (3 m/sec)	98.3* (4.5)	86.1 (3.7)
Two-coil arrangement (4 m/sec, upper or lower)	93.8 (4.7)	90.2 (3.2)
Four-coil arrangement (4 m/sec)	98.3* (2.9)	88.1 (5.2)

<sup>\*</sup> For these comparisons, the four-coil combination yielded a significantly higher average than the two-coil setup (P values are given in the text).

**Separation efficiencies at water velocities of 3 versus 4 m/second--**All sizes of test fish (fork lengths of 150-225 mm) were observed swimming upstream in the 3-m/second flow, while only the larger coho salmon were observed swimming for long in the 4-m/second flow. Although not statistically significant, average *SE*s were approximately 2% higher at 4 m/second than at 3 m/second whether the comparison was made for two or four coils (Tables 3 and 4). The higher water velocity created more turbulence within the rectangular flume, which appeared to cause fish to swim more actively to correct for the turbulence. Smaller fish could not swim for long, but larger coho salmon would unpredictably hold in the flume long enough to affect the *SEs*, just as they did in water velocities of 3 m/second. Exchanging the rectangular flume for a pipe or round-bottom flume should reduce the turbulent water conditions and therefore help improve the separation at 4 m/second.

Separation efficiencies for two distances between the last coil and diversion gate--The shorter distance between the lower interrogation unit and slide gate yielded slightly higher SEs than the longer distance between the upper interrogation unit and the gate, but the increase in SEs was not significant at 3 m/second (P = 0.381) or at 4 m/second (P = 0.805; Table 4). However, if only those tags that were targeted to be diverted are considered, one can calculate a diversion efficiency by combining the REs and SEs (DE = percentage of the tags read that were programmed to be diverted and were successfully diverted). The calculated DEs show that programmed fish that were read were separated significantly better over the shorter distance at both 3 m/second (P = 0.003) and 4 m/second (P = 0.033; Table 4). At both velocities, DEs were approximately 90% for the upper interrogation unit and close to 97% for the lower interrogation unit.

The 1.7- and 3.3-m distances between the last coil of the two interrogation units and the slide gate in this study are fairly typical of distances found at CRB dams. Although *SE*s were not significantly improved with the shorter 1.7-m distance, the significant improvement in *DE*s was dramatic. The *DE*s for the lower two-coil and four-coil arrangements were similar (all around 97%). Therefore, NMFS recommends that for future PIT-tag installations, diversion gates be installed at around 1 m (maximally 2 m) from the last interrogation coil. This would permit a higher percentage of PIT-tagged fish to be successfully diverted.

## **Conclusions and Recommendations**

Results for stick and fish trials using the four-coil arrangement at 4 m/second demonstrated that the *RE* and *SE* performance for double-read firmware was equivalent to the performance of the single-read firmware. Furthermore, the double-read firmware did not produce a single erroneous tag code. Thus, to avoid the potentially harmful erroneous tag codes, NMFS supports incorporating double-read firmware into the interrogation systems at the CRB dams. However, after NMFS finished its tests, Destron-Fearing

Table 4. Overall average separation efficiencies (*SE*s) and diversion efficiencies (*DE*s) for the two distances from the last coil to the diversion gate. Standard deviations are shown in parentheses. Groups were statistically compared using t-tests.

	Overall SE (%)	Overall DE <sup>a</sup> (%)
Two-coil arrangement(3m/sec, upper 2 coils)	86.8 (6.2)	88.1 (5.9)
Two-coil arrangement(3m/sec, lower 2 coils)	89.1 (4.1)	96.9 <sup>b</sup> (3.8)
Two-coil arrangement(4 m/sec, upper 2 coils)	89.9 (3.7)	92.7 (3.1)
Two-coil arrangement(4 m/sec, lower 2 coils)	90.5 (2.8)	96.6 <sup>b</sup> (3.2)

<sup>&</sup>lt;sup>a</sup> DE = (RE\*(SE/100)).

b In these comparisons, the lower 2 coils yielded a significantly higher average than the upper 2 coils (P values are given in the text).

produced a new generation of 400-kHz tags that incorporated the more accurate CRC method for error checking. Therefore, PSMFC will install these CRC firmware chips into PIT-tag interrogation equipment for the 1996 juvenile outmigration.

Increasing the number of interrogation coils from two to four coils significantly improved the ability to detect fish. At 3 m/second, average RE for the four-coil arrangement (98.3  $\pm$  4.5%) was significantly higher (P = 0.024) than average RE for the two-coil arrangement (93.6  $\pm$  7.3%). At 4 m/second, average RE for the four-coil arrangement (98.3  $\pm$  2.9%) was also significantly higher (P < 0.001) than average RE for the two-coil arrangement (93.8  $\pm$  4.7%).

Although not statistically significant, average *SE*s were approximately 2% higher at 4 m/second than at 3 m/second whether the comparison was made for two or four coils. The 4-m/second water velocity created more turbulence within the rectangular flume than did the 3-m/second water velocity. This greater turbulence appeared to cause the fish to swim more actively to correct for the turbulence. The smaller fish could not swim for long, but the larger coho salmon would unpredictably hold in the flume long enough to affect the *SE*s just as they did in water velocities of 3 m/second. Exchanging the rectangular flume for a pipe or round-bottom flume should reduce the turbulent water conditions and therefore help improve separation at 4 m/second.

The shorter distance between the lower interrogation unit and slide gate yielded slightly higher SEs than the longer distance between the upper interrogation unit and the gate, but the increase in SEs was not significant at 3 m/second (P = 0.381) or at 4 m/second (P = 0.805). Calculated DEs showed that the programmed fish that were read were separated significantly better over the shorter distance at both 3 m/second (P = 0.003) and 4 m/second (P = 0.033). At both velocities, DEs were <90% for the upper interrogation unit and close to 97% for the lower interrogation unit.

Therefore, NMFS recommends that for future PIT-tag installations, diversion gates be installed at around 1 m (maximally 2 m) from the last coil. This would permit a higher percentage of PIT-tagged fish to be successfully diverted.

# **Evaluation of Three Generations of 400-kHz Transponders**

#### Introduction

The 400-kHz PIT tags used throughout the CRB are purchased from Destron-Fearing Inc. All of the PIT tags used to tag fish before 1995 contained Atmill computer chips. When Atmill computer chips became unavailable, Destron-Fearing converted to Eurocell chips for their production tags. Tags containing Atmill computer chips were designated as Generation-1 PIT tags and those with Eurocell chips as Generation-2 PIT tags. As previously explained, these Generation-2 PIT tags were also different because they contained CRC error checking.

Generation-2 PIT tags were delivered to the CRB fisheries community for the 1995 season, but they were not evaluated before they were delivered. Soon after salmon started to migrate through the CRB bypass/collection facilities in 1995, PSMFC personnel observed that PIT-tag reading efficiencies for Generation-2 tags were significantly less than those for Generation-1 tags. The fisheries community sought to find out why and to determine if anything could be done immediately to improve the reading efficiencies. NMFS electronic engineers investigated and discovered that the return signals for Generation-2 tags were one-third less than for Generation-1 tags. This lower return signal would explain why some Generation-2 tags might not be read in the electronically noisy environments at the dams.

Destron-Fearing then determined that by modifying the receive circuitry in the exciter boards, electronic noise affecting the return signal would be reduced. This meant that to improve reading efficiencies at the dams, each exciter board (one per coil) throughout the entire CRB had to be modified after the migration season had begun. In addition, laptops running the BYCODE computer program and some necessary hardware were installed so that the slide gates could be triggered using all four coils instead of the normal setup that used only the two lower coils. All of the modifications were completed before the peak migration period; however, some data were obviously lost during the weeks before the modifications were in place.

The exciter modifications did increase the reading efficiencies of Generation-2 tags; however, even after the changes, reading efficiencies during the 1995 outmigration season were lower for Generation-2 tags than for Generation-1 tags (Carter Stein, unpubl. data, PSMFC, 45 SE 82nd Dr., Suite 100, Gladstone, Oregon 97027-2522). The discrepancy between reading efficiencies was < 5% for individual coils that have traditionally yielded reading efficiencies above 90%, but for coils with reading efficiencies normally below 85%, the median discrepancy was closer to 15%. This suggested that Generation-2 tags were less likely to be read under marginal conditions, such as where turbulence causes poor fish orientation.

An additional reason for the lower reading efficiencies observed might be that the high excitation levels maintained at the dams were turning off the computer chips in the

Generation-2 tags. The improved silicon in the computer chips means that Generation-2 tags require less power to energize them than Generation-1 tags. This means they turn on (become active) farther away from a coil, but it also means that a lower level of high power is necessary to turn them off. Therefore, it might be possible that a Generation-2 tag would be turned on as it approached a coil, but before its weaker return signal could be decoded, the tag would be turned off when it entered the stronger electromagnetic field within the actual coil. This potential cause was not examined during the 1995 season.

In an attempt to return the performance of their 400-kHz PIT tags to Generation-1 levels, Destron-Fearing switched to Hughes Microelectronics computer chips (Generation-3 tags) in September 1995. In order to avoid the in-season problems experienced in 1995, BPA asked NMFS to evaluate the Generation-3 tags before PSMFC bought them. Performance of all three generations of tags was compared using the PIT-tag test facility at NMFS Manchester Research Station. Effects of tag orientation (to simulate marginal reading conditions) and different excitation levels were examined.

#### **Methods and Materials**

The 10-cm- and 25-cm-diameter pipe sections at the PIT-tag test facility at Manchester (see Fig. 8) were used for this tag evaluation. Four interrogation coils are installed on each pipe. During testing, water velocity was maintained at approximately 3 m/second. Although no tag separation was done, the tag-reading data were recorded using the BYCODE computer program. A new computer file was generated for each replicate during the evaluation. To evaluate the tags, 15 tags from each generation were used. The tags were inserted into 15-cm wooden sticks whose ends were drilled to keep the tags securely in either optimal 0° orientation (tags inserted parallel to the long axis of the stick) or in marginal orientation (tags inserted at 45° angles to the long axis). For each replicate, all tags were either inserted at 0° orientation or at 45° orientation. Sticks were introduced individually into the pipes at intervals of 2-3 seconds.

**Orientation--**The 10-cm-diameter pipe was used in the evaluation of effects of orientation on reading efficiency because its narrow size kept the floating wooden sticks perpendicular to the coils so that tag orientation would not change during a test. The 15 tags from the three generations were fed through the pipe 10 times in both orientations. During these tests, excitation power levels were maintained at the 1.00-A setting, which is the standard level for the CRB.

**Excitation level--**The 25-cm-diameter pipe was used in the evaluation of effects of excitation level on reading efficiency. The larger pipe allowed the wooden sticks to rotate slightly from side to side in the flowing water, and thus more closely simulated fish passage through PIT-tag interrogation systems. Only tags in the optimal 0° orientation were used. Three excitation power settings were examined: 1.00 (normal level), 0.75, and 0.55 A. Twenty replicates were run at 1.00 A, 10 replicates at 0.75 A, and 6 replicates at 0.55 A.

**Statistics--**At the end of each replicate, the computer data file was analyzed to determine individual PIT-tag interrogation coil reading efficiencies. The number-of-coils-read/tag was also generated (maximum was 4 coils/tag). These numbers were then used in one-way analyses of variance (ANOVAs) to compare the effects of orientation and excitation power level on the three generations of tags. The significance level was established at  $P \le 0.05$ . Significant F values were further analyzed with Tukey tests.

#### **Results and Discussion**

**Orientation--**With tags in the optimal 0° orientation, none of the Generation-1 tags was missed by an interrogation coil, while both Generation-2 and Generation-3 tags were occasionally missed by one coil during a replicate (Table 5). However, the resulting number-of-coils-read/tag averages for each generation (4.00, 3.95, and 3.95 for Generations 1, 2, and 3, respectively) were not significantly different (P = 0.090). In contrast, when the tags were tested at the 45° orientation, no Generation-3 tags and only one Generation-2 tag were read by all 4 coils in all 10 replicates, while most of the Generation-1 tags were read by all 4 coils. The resulting number-of-coils-read/tag averages for each generation were significantly different (P < 0.001; Table 5). A Tukey test separated the Generation-1 average (3.95 number-of-coils-read/tag) from those of the other two generations (3.13 and 3.09 number-of-coils-read/tag for Generation-2 and Generation-3 tags, respectively).

These results supported the contention that poor tag orientation combined with reduced return-signal strength were significant causes for the lower reading efficiencies by Generation-2 tags within the CRB during the 1995 season. They also suggested that under normal monitoring conditions, Generation-3 tags would not be an improvement over Generation-2 tags and that a further decrease in tag reading efficiency could be expected.

**Excitation level--**If the poor performance observed in the 10-cm pipe was from high excitation power levels, then Generation-2 and Generation-3 tags should have done better at lower exciter settings. However, results from the excitation level evaluation indicated that performance of Generation-2 tags did not change over the three exciter power settings (P = 0.335; Table 6). The number-of-coils-read/tag averages for Generation-2 tags were 3.56, 3.56, and 3.70 for 1.00-, 0.75-, and 0.55-A settings, respectively. Although the performance of Generation-3 tags was significantly different at the three settings (P < 0.001), it did not follow a logical sequence. The number-of-coils-read/tag average was lowest at the 0.75-A setting ( $\overline{\times} = 2.53$ ) and highest at 1.00 A ( $\overline{\times} = 3.25$ ). In fact for some unknown reason, 11 tags out of 150 tags were completely missed at the 0.75-A setting. The Tukey test indicated that the 0.75-A average was significantly different from the 1.00-A and 0.55-A averages for Generation-3 tags.

Table 5. Number-of-coils-read/tag averages are presented for the three generations of tags from the tag-orientation test. Fifteen tags were used in 10 replicates to generate each average. Standard deviations are shown in parentheses. P values are from one-way ANOVAs. Superscript letters are used to distinguish significantly distinct groupings from a Tukey test.

Tag orientation	Gen. 1	Gen. 2	Gen. 3	P value
0° Orientation				
Average SD	4.00(0.00)	3.95(0.23)	3.95(0.23)	0.090
45° Orientation				
Average SD	3.95 <sup>a</sup> (0.22)	3.13 <sup>b</sup> (0.76)	$3.09^{b}(0.73)$	< 0.001

Table 6. Number-of-coils-read/tag averages are presented for the three generations of tags from the excitation level test. Fifteen tags were used in 10 replicates to generate each average. Standard deviations are shown in parentheses. P values are from one-way ANOVAs. Superscript letters are used to distinguish significantly distinct groupings from Tukey tests among the generations and superscript numbers for results from the Tukey test analyzing the significant within-generation ANOVA.

Excitation level	Gen. 1	Gen. 1 Gen. 2		P value
1.00 Amp				
Average SD	3.79 <sup>a</sup> (0.54)	$3.56^{b} (0.83)$	$3.25^{c,1}(0.98)$	< 0.001
0.75 Amp				
Average SD	3.86 <sup>a</sup> (0.46)	$3.56^{b} (0.85)$	2.53 <sup>c,2</sup> (1.28)	< 0.001
0.55 Amp				
Average SD	3.79 <sup>a</sup> (0.68)	$3.70^a (0.71)$	$3.02^{b,1} (0.99)$	< 0.001
P value-within generation	0.436	0.335	< 0.001	

### **Conclusions and Recommendations**

The overall results strongly indicate that Generation-1 tags performed significantly better than both Generation-2 and Generation-3 tags, as they had significantly higher number-of-coils-read/tag averages when the tags were in marginal orientation and at all three exciter power settings (Tables 5 and 6). Results from excitation tests proved that the poor performance by the Generation-2 and Generation-3 tags was not from being turned off by high excitation power levels. In comparing Generation-2 and Generation-3 tags only, Generation-2 tags had significantly higher averages than Generation-3 tags at all three exciter power settings. These results suggest that under normal monitoring conditions, Generation-3 tags would not be an improvement over Generation-2 tags. Destron-Fearing is working on another modification to the Generation-3 tag (increasing its signal modulation). This change was not made before PSMFC's ordering deadline of December 1995 and consequently, based on the above results, PSMFC ordered Generation-2 tags for the 1996 season.

# **Evaluation of Generation-3B PIT Tags**

#### Introduction

Since the fisheries community would prefer to buy tags that match the performance of Generation-1 tags, NMFS recommended that the modified Generation-3 tags (Generation-3B tags) be tested when they were produced by Destron-Fearing. Destron-Fearing brought Generation-3B tags to Manchester in February 1996 when the following evaluation was performed.

#### **Methods and Materials**

In this evaluation, tag orientation tests were run in both the 10-cm and 25-cm pipe sections. Thirty-five-cm sticks were used, which allowed tags to be inserted on both ends. Thus, tags were inserted at 0° orientation on one end and 45° orientation on the other. Instead of 15, only 10 tags from each generation were tested in each orientation. The full complement of 20 Generation-3A tags was not available and so only 10 tags inserted at 45° were used. Ten replicates were run in each pipe.

The number-of-coils-read/tag numbers were used to run one-way ANOVAs to compare the effects of orientation on the three generations of tags. The significance level was established at  $P \le 0.05$ . Significant F values were further analyzed with Tukey tests.

#### **Results and Discussion**

In the 10-cm pipe with tags in the optimal  $0^{\circ}$  orientation, none of the Generation-1 tags was missed by an interrogation coil, while only two tags were missed by interrogation coils for Generation-3B tags. In contrast, at least one Generation-2 tag was missed by one coil in every replicate. Consequently, the resulting number-of-coils-read/tag averages for each generation (4.00, 3.71, and 3.98 for Generations 1, 2, and 3B, respectively) were significantly different (P < 0.001; Table 7). A Tukey test separated the Generation-1 and Generation-3B tags from the Generation-2 tags.

In the 10-cm pipe with tags in the marginal  $45^{\circ}$  orientation, more tags were missed by all of the generations. The number-of-coils-read/tag averages for each generation were significantly different (P < 0.001) among generations (Table 7). A Tukey test separated the Generation-1 and Generation-3B averages (3.78 and 3.85 number-of-coils-read/tag for Generation-1 and Generation-3B tags, respectively) from the other two generations (2.75 and 2.77 number-of-coils-read/tag for Generation-2 and Generation-3A tags, respectively).

Table 7. Number-of-coils-read/tag averages are presented for the three generations of tags from the tag orientation tests performed in the two pipes. Ten tags were used in 10 replicates to generate each average. Standard deviations are shown in parentheses. P values are from one-way ANOVAs. Superscript letters are used to distinguish significantly distinct groupings from Tukey tests among the generations.

		0° orientation	45° orientation
10-cm pipe		·	-
Generation 1			
Average	SD	$4.00^{a}(0.00)$	$3.78^{a}(0.42)$
Generation 2			
Average	SD	$3.71^{b}(0.48)$	2.75 <sup>b</sup> (0.67)
Generation 3A	<b>L</b>		
Average	SD		2.77 <sup>b</sup> (0.66)
Generation 3B	}		
Average	SD	$3.98^{a}(0.14)$	$3.85^{a}(0.36)$
P value		< 0.001	< 0.001
25-cm pipe			
Generation 1			
Average	SD	4.00(0.00)	$3.90^{a}(0.36)$
Generation 2			
Average	SD	4.00(0.00)	3.21 <sup>b</sup> (1.23)
Generation 3A	<b>L</b>		
Average	SD		2.42°(1.22)
Generation 3B	}		
Average	SD	3.97(0.17)	$3.87^{a}(0.42)$
P value		0.381	< 0.001

In the 25-cm pipe, with tags in the optimal  $0^{\circ}$  orientation, almost no tags were missed by any interrogation coils and the ANOVA showed no statistically significant differences (P = 0.381; Table 7). With tags in the marginal 45° orientation, tags were completely missed by all four coils for both Generation-2 and Generation-3A tags while none of the Generation-1 and Generation-3B tags was missed completely. The ANOVA showed statistically significant differences (P < 0.001), and the subsequent Tukey test separated the four groups into three groupings: 1) Generation-1 and Generation-3B tags, 2) Generation-2 tags, and 3) Generation-3A tags.

## **Conclusions and Recommendations**

The results from this February 1996 evaluation strongly suggest that under normal monitoring conditions, these Generation-3B tags will perform as well as the original Generation-1 tags. Therefore, NMFS recommends that the fisheries community use these Generation-3B tags. Unfortunately, these tags were not available for the 1996 spring tagging season, but were for the summer and fall tagging seasons.

# Toxicity Evaluation of the Dye used to Detect Broken PIT-tag Casings

#### Introduction

PIT tags are subjected to a series of quality-control tests during their manufacture. Pressure tests are conducted to detect cracks or damage in the glass that encapsulates the tags. For these pressure tests, the newly produced tags are placed in a container with a dye and pressurized at 413.7 kPa (60 psi) for 2 hours. During this time, the dye penetrates broken tags and makes them easy to identify. After this exposure the tags are removed, air dried thoroughly, and the broken ones are rejected.

In 1993, Destron-Fearing switched their tag manufacturing to Hughes Microelectronics in Spain. Hughes Microelectronics uses a green dye produced in Spain in its pressure tests. At one time in the 1980s, Destron Inc. (Previous name of Destron-Fearing) used a red dye that NMFS subsequently determined was lethal to fish. They immediately discontinued its use after NMFS notified them of the problem. Because of this past problem, a 72-hour survival study was conducted with the green dye to determine whether or not it was toxic to fish.

## **Methods and Materials**

The test dye, mint green dye #1732, is manufactured by Aromas Maluquer SA and it contains American Food Yellow 5 and Food Blue 5 in addition to some proprietary ingredients. To start our evaluation, one batch of PIT tags was soaked in the test dye (70 ppm) and a second batch soaked in 100% ethanol for 72 hours. All of the PIT tags were air dried for 2 hours prior to use.

On 3 October 1994, presmolt coho salmon were randomly divided into four groups of 30 fish: 1) those injected with regular PIT tags that had been soaked in ethanol, 2) those injected with dyed PIT tags, 3) those injected with 0.5 mL of dye, and 4) those fin-clipped that represented controls. All fish were anesthetized with Tricaine Methanesulfonate (MS-222) before being handled. Group 3 was injected intraperitoneally with 0.5 mL of the dye using an automatic dispenser and a 27-gauge needle. Groups 1 and 2 were PIT-tagged using the procedure described by Prentice et al. (1990b) and Group 4 was fin-clipped using standard procedures. Fork lengths were measured to the nearest millimeter and weights were taken to the closest 0.1 g on 10 fish from each group. All groups were held for 72 hours in a 1.2-m circular tank and monitored for survival and unusual behavior.

One-way ANOVAs were used to compare fork lengths and weights of the four groups at the time they were tagged. Statistical significance was set at  $P \le 0.05$ . Since no mortality occurred during the test, no statistics were conducted on the survival data.

## **Results and Discussion**

When the fish were tagged, there was no significant difference in fork lengths (P=0.259) or weights (P=0.451) among the four groups (Table 8). There were no mortalities during the 72-hour observation period and fish behavior was normal.

# **Conclusions and Recommendations**

Since the dye (mint green dye #1732) does not appear to be lethal to fish or cause abnormal behavior, NMFS concludes that it is an acceptable dye for the pressure-testing procedure.

Table 8. Average fork lengths and weights of 10 individuals from the four groups of coho salmon at the time of tagging. Standard deviations are shown in parentheses. P values are from one-way ANOVAs.

	Regular PIT tags	Dyed PIT tags	Injected dye	Control	P value
Weight (g)					
Average SD	12.4	13.1	11.5	13.1	0.259
	(2.5)	(2.6)	(3.1)	(1.9)	
Fork length (mm)					
Average SD	103.8	109.0	104.7	111.3	0.451
	(7.9)	(12.3)	(10.3)	(6.7)	

# Electromagnetic Field Effects on Reproducing Fish: Medaka (*Oryzias latipes*)

#### Introduction

PIT-tag interrogation systems that monitor juvenile and adult salmon as they move through bypass/collection facilities at CRB Dams are an integral part of the PIT-tag program. The PIT-tag interrogation units currently used to monitor migrating salmon operate at 400 kHz. In the future, operating frequencies between 120 and 135 kHz will have to be used if the fisheries community is to reach its goal of interrogating returning adult salmon in fish ladders (Prentice et al. 1993, Prentice et al. 1994). In 1989, NMFS started investigating several 400-kHz PIT-tag interrogation units for monitoring the volitional movement of juvenile and adult salmon as they migrated within streams and into and out of hatcheries. During the studies evaluating adult salmon passage through interrogation units at Minter Creek and Skagit River Washington State Hatcheries, biologists observed that volitionally migrating adults remained within the interrogation units for an average of 2 minutes, but that some fish remained for several hours (Prentice et al. 1994). The potential for long exposure of migrating adult salmon to strong electromagnetic fields (EMFs) within interrogation units caused concern among NMFS personnel.

Studies by others have documented that EMFs in both kHz and GHz ranges can produce negative biological effects under prolonged (months) exposure (see reviews by Aldrich and Easterly 1987, Brown and Chattopadhyay 1988). Regardless of the operating frequency used, even the weakest calculated field strength within a PIT-tag interrogation unit (58 A/m for 5,551 cm² passageways) is substantially higher than the 1.6 A/m level permitted under 1982 American National Standards Institute (ANSI) standards for 24-hour exposures to an entire human body by EMFs in the 100-to-400-kHz range.

Unfortunately, no studies have investigated the effects of EMFs in the 100-to-400-kHz range on the biology of animals. Therefore, prudence dictated that NMFS determine if there were any negative impacts on the reproductive success of fish before interrogation units for adult salmon were installed on a wide scale both within and outside of the CRB. NMFS designed two studies to investigate whether there were any biological effects from the types of exposure adult salmon were likely to face. Since adult salmon die after spawning, the concern was more for their offspring and subsequent generations than for the adults physically exposed.

<sup>&</sup>lt;sup>1</sup> In 1996, the decision was made to base the next PIT-tag interrogation system for the CRB on the 134.2-kHz standard approved by the International Standard Organization. The interrogation units for juvenile salmon will be installed for the Year 2000 outmigration season. Development of interrogation units for adult salmon is on-going.

From 1990 through 1993, NMFS conducted two studies to determine if fish or their offspring were affected by exposures up to 24 hours to 125-kHz or 400-kHz EMFs (Prentice et al. 1994). In the study that exposed chum salmon (*Oncorhynchus keta*) zygotes directly, no significant differences or trends were found in the number of survivors, average fork lengths, or percent deformities between 24-hour exposed and unexposed groups. In the other study, medaka (*Oryzias latipes*) were used as a surrogate species for salmon. Medaka, freshwater killifish, were chosen for their relatively short generational time (4-6 months), ability to reproduce year-round, common use in teratological studies, and being oviparous like salmonids.

In this medaka study, actively breeding fish were exposed to a range of times (1-1400 minutes) under significantly stronger EMFs (4-5 times) than would be present within an interrogation unit for adult salmon. NMFS reasoned that if no impact was documented on reproduction or development over two generations, then we could assume that shorter and weaker exposures would not negatively affect other species (e.g., salmon). However, if any of these exposures affected medaka, then more study would be needed.

In the first-generation medaka offspring, there were differences in larval mortality between the control (20.1%) and EMF-exposed groups (27.3-33.7%). In addition, the control group had fewer deformed hatched larvae (3.0%) than the EMF-exposed groups (5.0-11.5%). Although large, these differences were not significant because statistical power was low with only six replicates completed. However, the results did suggest that EMF exposure may affect the survival and performance of first-generation offspring from EMF-exposed fish.

Since the data from second-generation fish indicated no differences in performance between the offspring from control and exposed fish, a modified experimental design was implemented in 1994 to concentrate on evaluating first-generation offspring performance through the transition to exogenous feeding. It was vital to include this period of transition to exogenous feeding because other research studies have found this transition to be a critical period when "treated" fish have exhibited significantly higher mortalities or abnormalities than untreated controls (e.g., Rand and Petrocelli 1985, Blaxter 1988). This modified experimental design would also permit enough replicates (10) to be accomplished to provide the necessary statistical power for determining whether trends like those listed above are significant or merely due to normal biological variation. The modified experimental design also expanded on the first study to test not only tag-energizing frequency, but also field strength and field orientation. This report covers this second medaka experiment.

If this study indicated there were significant negative effects, then the next step would be to determine if interrogation units for adult salmon could be designed that would reduce the EMF exposure to an acceptable level.

### **Methods and Materials**

In this cooperative study with the University of Washington, actively breeding medaka were exposed to 12 treatments to test tag-energizing frequency, field strength, and field orientation (Table 9). The control was duplicated to give a better estimate of the within-species variation (i.e., normal biological variation for this species). As indicated in Table 9, we used capital letters to designate the 12 treatments.

To conduct this study, fish culture and EMF-exposure laboratories were constructed at the NMFS Manchester Research Station. To induce egg production in medaka, the same temperature (25-27°C) and light conditions (16 hours light and 8 hours dark) were maintained in both laboratories. For exposing the fish, personnel from the NMFS Sand Point Electronics Shop built four exposure units (52-cm long by 25-cm wide by 30-cm high) from Plexiglas. Each had a single coil wrapped around it that consisted of 26 wraps of 18-gauge Litz wire. Three of the exposure units had horizontal coils and one had a vertical coil. One of the horizontal exposure units operated at 400 kHz while the others operated at 125 kHz. Different settings on the power amplifiers were used to produce the two field strengths tested: approximately 50 and 260 A/m at the centers of the exposure units. The applied 10-A, peak-to-peak current was continuous, not pulsed. For the control treatments, no current was applied to an exposure unit.

Since multiple treatments were to be conducted simultaneously in the EMF-exposure laboratory, NMFS contracted Pacific Northwest National Laboratory (PNNL) to take EMF measurements in the laboratories using equipment calibrated in frequency ranges appropriate for the both the ELF (extremely low frequency) and RF fields involved. PNNL's measurements affirmed that the exposure units did not interfere with each other when they were placed 2.4 m apart. Their measurements also confirmed the calculated magnetic field flux density distributions within the exposure units. In addition, they determined that the minimal background (60-300 Hz range) magnetic fields in the two laboratories, as well as those within the electric incubator, would not interfere with the higher RF exposures used in the study.

Medaka were cultured under static water conditions following the methods of Kirchen and West (1976). Broodstock from Japan were used in this study and were purchased through local tropical fish stores. Although Kirchen and West had success rearing medaka year-round, we and other researchers contacted found the fish were primarily dormant during the winter months. We also found broodstock procurement was inconsistent over the entire study because shipments were delayed, and diseases, primarily ick, caused problems. In fact, the last three shipments did not survive the 10-14 day quarantines at the fish store. This helped us in deciding to stop the study before the tenth replicate was fully completed because winter was arriving when the study would be shut down for 3-4 months. Furthermore, the stop decision took into account that the statistical results had been the same since the sixth replicate.

Table 9. Coil orientation, tag-energizing frequencies (kHz), field strengths (A/m) and time exposures (minutes) for the 12 treatments. In addition, the letters used to designate each treatment are presented.

Coil orientation	Frequency (kHz)	Field strength(A/m)	Time exposure (minutes)				
No field Horizontal	400	260	260 0* -1		-140	-1,400	
Horizontal	125	260	-	1	140	1,400	
Vertical	125	260	-	-	-	1,400	
Horizontal	125	50	-	1	140	1,400	
			Treatment abbreviations				
			A, B	-C	-D	<b>-</b> E	
			-	F	G	Н	
			-	-	-	I	
			-	J	K	L	

<sup>\*</sup> The control was duplicated (i.e., treatments A&B) to give a better estimate of within-species variation or normal biological variation.

After the broodstock had successfully completed their quarantine at a fish store, the fish were transported to our laboratory. Upon arrival, broodstock were maintained separately for 3-5 days to get the fish acclimatized to the new conditions and to confirm that they were healthy. The individual fish were then sexed to stock the 19-L aquariums with sets of 9 females and 6 males. Typically, 15-20 sets of fish were maintained at a time. The aquariums were kept bare except for a sponge filter. The fish were transferred to clean aquariums 2-3 times/week.

Fish were maintained in an aquarium until at least 3 out of 9 females were brooding on a daily basis before the set was considered ready for exposure. Female medaka produce external clutches of eggs that remain attached to their abdomens for 4-6 hours after fertilization until the adhesive material that binds the eggs together begins to disintegrate. At this time, the females tend to rub the eggs off of themselves. Loose dangling eggs are also eaten by other fish. Thus, eggs attached to females represented only the current day's production and not multiple days' production. This made it possible to collect only eggs that had been produced while the fish were being exposed and the two subsequent days. Larval development is also rapid and easily followed in the clear eggs, which made it easy to confirm the age of the eggs.

Three sets of medaka were exposed at one time. Therefore, when fish from three aquariums were actively breeding, each aquarium was randomly assigned to one of the 12 treatments until that replicate was completed. To perform an exposure, medaka were transported in their aquariums to the EMF-exposure laboratory. There, they were positioned in the centers of the exposure units. To help keep the treatments unknown to the main investigator, programmable timers were connected to the power amplifiers and aquarium labels only included the exposure number (e.g., the first aquarium used in the second replicate would be labeled 13). All of the treatments began at 1100 hours, and each of the 12 treatment groups remained in an exposure unit for 1,400 minutes, regardless of how long the induced EMF was present. The fish were then transported back to the culture room for egg collection.

Clutches of eggs were collected from all breeding females on the morning an aquarium was transported back to the culture room and for the next 2 days. To collect eggs, individual females were netted and eggs gently removed from her abdomen. The number of eggs produced by each female was recorded. On each day, up to 25 eggs from one aquarium were all placed into a single 4-cm-diameter petri dish containing a liquid saline growth medium recommended by Kirchen and West (1976). If > 25 eggs were collected, then two or more dishes were used. On the first day, eggs were also collected from the bottom of the aquarium and maintained in separate petri dishes. Any older eggs (easily identified by egg development) were discarded. This extra step was necessary because eggs were sometimes dislodged during the short drive back from the EMF-exposure laboratory. At the end of the first day, the fish were transferred to clean aquariums.

Eggs collected over the 3 days and the number of breeding females were combined to determine the average number of eggs produced per female for that treatment (the eggs collected from the bottom of the aquariums were omitted from this calculation). On the day the eggs were collected, they were inspected for fertilization. From this information, fertilization rates (number of fertilized eggs/total number of eggs) were calculated for each treatment. After the third day, the adults were sacrificed and measured to the nearest 0.1 mm using a customized measuring slide under a dissecting microscope.

The petri dishes were placed into a 24°C electric incubator and the offspring were examined daily until all had either died or hatched (hatching starts at around Day 14). During these daily examinations, unfertilized eggs, dead eggs, dead larvae (developing and hatched), and grossly deformed hatched larvae that obviously would not survive were counted, removed from petri dishes, and preserved in Bouins solution. If fungus was observed during the daily examinations, then precautionary measures were taken because fungus easily spreads even to healthy eggs. Treatments that had eggs with even slight cases of fungal infestation were separated into new petri dishes; one for the infected eggs and one for the uninfected eggs. However, this approach was not always successful in halting the fungal spread in the supposedly uninfected group. Growth medium in the petri dishes was minimally replaced twice a week (more often when fungus was present).

Hatched larvae that were active and had normal morphologies were immediately transferred to the juvenile-rearing tanks. Inactive hatched larvae were left in the petri dish until they either became active or died. The latter were recorded as deformed hatched larvae, as were active larvae that had curved spines or missing fins (these would die shortly after hatching and so were not transferred). Any eggs that had not hatched by one month following the exposure date were also recorded as deformed hatched larvae (all of these were followed initially, but even if they eventually hatched with normal shaped bodes, they never became active enough to be transferred to a juvenile tank). From these data, larval mortality (number of dead larvae/number of fertilized eggs) and deformity rates among the hatched larvae (number of deformed hatched larvae/total number of hatched larvae) were calculated.

Separate juvenile-rearing tanks were used to house juvenile medaka from each treatment. These rearing tanks were rectangular plastic containers that held 2 L of water. The water was maintained under static conditions (an air stone kept the water circulating) at approximately 24°C and was changed twice a week. The juvenile medaka were fed commercially prepared juvenile fish feed. Juvenile medaka were observed daily and any mortalities were removed and recorded. In addition, when the water was changed, the numbers of juveniles were counted.

Thirty days after half of the eggs in a treatment had hatched, all juveniles in that treatment were sacrificed (this ensured that the last hatching fish had passed through the transition to exogenous feeding). The fish sacrificed were used to yield estimates of

juvenile mortality (number of sacrificed juveniles/number of transferred hatched larvae). In addition, fork lengths of the sacrificed juveniles (a maximum of 30 juveniles/treatment) were measured to the nearest 0.1 mm using a compound microscope attached to a computer running Optimas, an image analysis program.

Broodstock size, egg production/female, fertilization, mortality, length, and deformity data for the 12 treatments were statistically analyzed with one-way ANOVAs. The significance level for all tests was established at  $P \le 0.05$ .

### **Results and Discussion**

The 9+ replicates were completed over a 2.5-year period, from May 1994 to October 1996. As indicated above, difficulty in broodstock procurement and inconsistent egg production both prolonged the study and helped lead to the decision to stop the study before the tenth replicate was completed. When the study was stopped, half of the treatments had nine replicates and half of them had ten replicates (Table 10). There was no difference in the size of broodstock among the 12 treatments (F = 1.087; P = 0.386; Table 10). Most of the fish measured between 27 and 31 mm.

Average egg production ranged from 4.3 to 5.9 eggs/female and was not significantly different among the 12 treatments (F = 0.355; P = 0.970; Table 10). Treatment K was the only treatment whose average was below 5.0 eggs/female, but all of the treatments had replicates in which its value was less than 3.5 eggs/female. There appears to be a naturally large variation in egg production for this species. For example, in Replicate 4, the averages for Control A and Control B were 4.4 eggs/female and 9.1 eggs/female, respectively (Table 11). Having a large number of treatment cells (18 for the controls compared to 6 in the first study and 93 for the exposed groups compared to 20) helped to confirm the large biological variation displayed by this species by showing that the range within a treatment is as large as between treatments. Such variation in individual female performance is well documented for other species including salmonids (e.g., Refstie and Gjerdem 1975, Blanc and Chevassus 1979). When averaged over 9+ replicates, means for the control groups (5.5 eggs/female) and the EMF-exposed groups (5.4 eggs/treatment) indicated that egg production was basically the same whether the breeding medaka were exposed or not.

There was no significant difference in the percentage of eggs fertilized among the 12 treatments (F = 0.842; P = 0.599; Table 10). Average fertilization rates for the 12 treatments ranged from 87.9 to 96.0%. Treatment C was the only treatment whose average was below 90.0%. It was low due to two females; one had five out of its seven eggs unfertilized and the other had all eight of its eggs unfertilized. The latter female's eggs were probably removed before a male had a chance to fertilize them. Without these two females, the fertilization percentage for Treatment C would have equaled 92.6%.

Larval mortality rates were not significantly different among the 12 treatment groups (F = 0.872; P = 0.570; Table 12). Averages for treatments in this study (7.2-17.9%) were less than averages in the first NMFS medaka study (20.1-33.7%; Prentice et al. 1994). In the first study, only 1 treatment cell out of 30 had less than 7% larval mortality, while in this study 55 out of 111 treatment cells or roughly 50% had larval mortalities < 7%. This is most likely because the petri dishes were checked 7 days/week instead of only 5 days/week. Therefore, if an egg died, it could be removed immediately, which reduced the opportunity for fungus to infest healthy eggs.

Fungus was present in all of the treatment cells that had > 20% larval mortality in this study. Fungus also appeared to be a factor in groups with high mortality rates in the first medaka study: 24/30 treatment cells had > 20% larval mortality and 22 of those 24 groups had fungus present. Similar to the other categories, there was a wide variation in larval mortality rates within each treatment whether it was a control or an EMF-exposed treatment. For example, for the two control treatments, larval mortality rates ranged from 1.5 to 32.1%. For Treatment I, which had the highest average at 18.1%, larval mortality rates ranged from 0.0 to 36.4%. Overall, average mortalities for control (13.1%) and EMF-exposed (10.8%) groups were similar in this study. Therefore, the worrisome trend observed in the first study was likely due to normal biological variation and some larval culture practices.

Percentages of deformed hatched larvae ranged from 2.2% to 11.2% and were not significantly different among the 12 treatment groups (F = 0.780; P = 0.659; Table 12). Except for Treatment C, whose average was 2.2%, all of the treatments had treatment cells with >12% and thus, there appeared to be a naturally large biological variation for this species. A genetic basis for this large biological variation in the number of deformed hatched larvae is supported by the fact that Replicates 7 and 8, which used the same broodstock, were responsible for 48% of the total deformed larvae observed. Average deformity rates for the control groups (5.9%) and for the EMF-exposed groups (5.2%) were higher than rates for the control group (3.0%) in the first medaka study (Prentice et al. 1994). However, if one compares identical EMF-exposed treatments between the first and second studies, Treatments D, E, and H all yielded 11.5% rates in the first study but only 5.8%, 3.7%, and 3.7% in this second study. Thus, unlike in the first study, there did not appear to be an increase in the percentage of deformed larvae in the EMF-exposed treatments.

Juvenile mortality rates were not significantly different among the 12 treatments (F = 1.082; P = 0.384; Table 12). Averages for juvenile mortality ranged widely, from 9.5% to 29.1%, over the 12 treatments. Similar to the situation with deformed hatched larvae, Replicates 7 and 8 were responsible for a large proportion (44%) of juveniles that died over the juvenile-rearing period. With averages for control fish (16.3%) and EMF-exposed medaka (18.2%) being similar, it appeared that each handled equally well the transition to exogenous feeding that has been found in other research studies to be a critical period when "treated" fish have exhibited significantly higher mortalities or abnormalities than untreated controls (e.g., Rand and Petrocelli 1985, Blaxter 1988).

Table 10. Summary results (averages and standard deviations) for the 12 treatments (see Table 9 for full description of treatments) that exposed actively breeding medaka to different EMF conditions. Eggs were collected over 3 days and cultured to a mid-juvenile stage. Probability values are based on one-way ANOVAs. At the bottom are the averages and standard deviations from combining the two controls and all of the exposed treatments.

Number of		Broodst	ock size	Eggs/	Eggs/female		ertilization
Treatment	replicates	Avg	SD	Avg	SD	Avg	SD
A	9	28.0	(2.5)	5.5	(2.2)	92.5	(8.9)
В	9	27.9	(2.6)	5.4	(2.1)	90.1	(15.3)
C	9	28.0	(2.4)	5.9	(2.6)	87.9	(13.3)
D	10	28.1	(2.2)	5.3	(2.4)	95.8	(5.4)
E	10	27.8	(2.0)	5.6	(1.5)	90.7	(7.3)
F	10	27.7	(1.8)	5.3	(3.1)	92.5	(7.8)
G	10	28.0	(2.6)	5.6	(1.4)	93.4	(6.4)
Н	9	27.5	(2.1)	5.6	(1.2)	93.4	(7.1)
I	10	27.6	(2.5)	5.4	(1.7)	95.5	(4.0)
J	9	27.9	(1.8)	5.0	(1.3)	92.3	(9.5)
K	9	27.7	(2.2)	4.3	(1.0)	96.0	(4.8)
L	10	28.1	(2.5)	5.7	(1.8)	95.8	(3.9)
P value			0.368		0.970		0.599
Controls	18	28.0	(2.6)	5.5	(2.1)	91.3	(12.2)
Exposed	96	27.8	(2.2)	5.4	(1.9)	93.4	(7.4)

Table 11. The eggs/female averages for all of the treatments for Replicates 4-7 showing the large amount of biological variation that is normal for medaka. Especially note the variation within replicates for the two control treatments (A&B).

Treatment	Replicate 4 Average	Replicate 5 Average	Replicate 6 Average	Replicate 7 Average
A	4.4	7.9	8.9	6.7
В	9.1	7.0	3.0	6.8
C	4.2	10.0	3.8	9.5
D	4.0	4.4	6.4	7.0
E	5.6	6.0	6.0	4.2
F	3.4	3.7	2.8	10.3
G	5.9	5.1	6.5	6.9
Н	5.5	7.4	4.3	5.0
I	7.3	4.3	8.1	5.7
J	6.7	2.8	6.0	5.6
K	2.7	4.5	5.3	4.1
L	5.3	7.1	6.4	6.9

Table 12. Summary results (averages and standard deviations) for the 12 treatments (see Table 9 for full description of treatments) that exposed actively breeding medaka to different EMF conditions. Eggs were collected over 3 days and cultured to a mid-juvenile stage. Probability values are based on one-way ANOVAs. At the bottom, are the averages and standard deviations from combining the two controls and all of the exposed treatments.

		La	ırval	Juv	enile	Pe	ercent				
	Number of	mo	rtality	moi	rtality	def	ormity	Overall	survival	Juvenil	e length
Treatment	replicates	Avg	SD	Avg	SD	Avg	SD	Avg	SD	Avg	SD
A	9	12.5	(9.8)	19.4	(11.3)	5.5	(5.1)	67.2	(14.5)	10.8	(2.1)
В	9	13.7	(10.0)	13.2	(14.7)	6.2	(6.5)	70.4	(16.8)	10.8	(2.0)
C	9	8.5	(5.1)	11.4	(11.0)	2.2	(2.7)	79.3	(11.5)	10.4	(2.0)
D	10	17.9	(17.0)	16.4	(16.9)	5.8	(9.8)	66.8	(26.3)	10.3	(1.4)
E	10	8.7	(6.5)	29.1	(22.4)	3.5	(3.7)	62.8	(20.2)	10.2	(2.0)
F	10	8.5	(9.3)	9.5	(9.0)	5.2	(7.1)	78.7	(14.3)	10.9	(2.2)
G	10	10.9	(21.6)	18.0	(16.9)	11.0	(10.1)	64.6	(22.4)	10.5	(1.3)
Н	9	7.2	(5.8)	17.6	(14.8)	3.7	(6.5)	72.2	(16.9)	10.0	(1.3)
I	10	15.6	(12.7)	22.2	(27.9)	4.5	(5.6)	61.3	(25.2)	10.8	(1.9)
J	9	10.7	(6.6)	22.0	(24.1)	7.0	(10.3)	65.9	(24.0)	10.8	(2.3)
K	9	8.6	(11.5)	24.2	(16.9)	4.1	(8.5)	68.8	(23.7)	10.7	(2.1)
L	10	10.5	(12.0)	13.2	(8.6)	5.0	(7.2)	74.8	(18.2)	10.1	(1.1)
P value		0.	570	0.	384	0	.659	0.	739	0.9	990
Controls	18	13.1	(9.6)	16.3	(13.1)	5.9	(5.7)	68.8	(15.3)	10.8	(2.0)
Exposed	96	10.8	(11.8)	18.2	(17.8)	5.2	(7.5)	69.6	(20.7)	10.5	(1.7)

Averages for fork lengths of preserved juveniles ranged from 10.0 to 10.9 mm and were not significantly different among the 12 treatments (F = 0.268; P = 0.990; Table 12). In the first replicates, some of the groups had smaller juveniles because they had higher numbers of juveniles. This density-dependent growth was eliminated by limiting the number of juveniles in each 2-L tank to 30 individuals. Thus, the lower averages for Treatments E, H, and L had to do with their having higher numbers of juveniles in early replicates, and were not due to these treatments being exposed to EMFs.

Overall survival rates from fertilization to the mid-juvenile stage ranged from 61.3 to 79.3% and were not significantly different among the 12 treatments (F = 0.697; P = 0.739; Table 12). As indicated by the large standard deviations, there was a naturally large variation in overall survival rates within a treatment. For example, Treatment B (a control), survival rates ranged from 47.1 to 90.9% for individual replicates. In Treatment D, which had the highest standard deviation, overall survival rates ranged from 33.3% to 100.0%. This wide variation is not surprising since the overall survival rates take into account most of the stages covered above that had displayed a large amount of natural variation for this species. Averages for the control groups (76.7%) and the EMF-exposed groups (78.5%) were similar, suggesting that EMF-exposure does not affect survival of this species.

## **Conclusions and Recommendations**

There were no significant differences between the control and the EMF-exposed treatments in any category (e.g., egg production/female, fertilization rates, larval mortality rates, deformity rates, overall survival). Duplicating the control treatment was critical for this study as the high standard-deviation values associated with the averages for the controls indicated that there is a large amount of natural biological variation in this species.

At this time, results suggest no negative effects from exposure to tested tag-energizing frequencies, field strengths, or field orientations. Assuming the results are directly transferable, they do not limit the design possibilities for developing adult salmon PIT-tag interrogation systems as long as the adults will not be exposed continuously for longer than 24 hours. Exposures longer than 24 hours might not be a problem, but the effects of this longer exposure would need to be tested if a design resulted in salmon being consistently exposed for >24 hours. Based on these results, NMFS recommends that the fisheries community continue toward its goal of PIT-tag interrogation of adult salmon in fish ladders.

# **PIT-Tag Retention in Adult Salmon**

#### Introduction

The PIT tag is a reliable tool for identification of juvenile and adult salmon. However, an earlier study showed that up to 40% of female and 20% of male coho salmon that were tagged as juveniles lost their tags during sexual maturation (Prentice et al. 1994). Tag loss was also observed in mature sockeye salmon reared in net-pens. Loss of PIT tags during sexual maturation limits their usefulness in situations where identification of mature adult fish is required (e.g., broodstock programs).

The PIT tag used in the Columbia River Basin is encapsulated in biologically inert glass and therefore it is usually found loose in the peritoneal cavity. PIT-tag manufacturers have found that by coating a tag with parylene or by adding a Teflon tip to the tag, they were able to stop PIT tags from migrating within small mammals. In this study, NMFS investigated whether these tags, as well as acid-etched regular PIT tags, would reduce tag movement and loss within fish. These tags were compared to unmodified or regular PIT tags for tissue response (e.g., encapsulation) and tag loss during sexual maturation. A group of fin-clipped, untagged fish was included as a control for comparing growth and mortality rates between tagged and untagged fish.

## **Methods and Materials**

**Experimental details/fish culture--**Juvenile coho salmon from Washington Department of Fisheries and Wildlife's Minter Creek Hatchery were transferred to the NMFS Manchester Research Station on 9 May 1995. Tagging was delayed when during the first weeks after transfer dead fish were found partially covered with fungus. Blocks of salt were added twice to the tanks to treat the fish. The fungus problem disappeared after this saline treatment.

The following designations were assigned to five treatment groups established in late June 1995 with 2,396 fish: "Capped" for PIT tags with Teflon tips, "Coated" for tags with parylene coating, "Etched" for tags etched with acid, "Regular" for unmodified tags, and "Clipped" for control fish that had their adipose fins clipped. Each fish was anesthetized with MS-222, tagged or clipped, its fork length measured to the nearest 1.0 mm, and its body weight taken to the nearest 0.1 g. After handling and tagging, fish from the five groups were evenly distributed into two 5.4-m-diameter tanks supplied with Beaver Creek water.

Originally, the experimental design called for the five groups to be subsampled each fall and spring until the last of the salmon reached maturity in late 1997. During each subsample, all fish would have their fork lengths measured for growth, all tag codes would be recorded to have an accurate record of the number of surviving fish in each group, and 100 fish from each group would be sacrificed to examine tissue response and measure body weight. However, when the fish population experienced high mortality

during late July, the first subsample was delayed until spring 1996 (Fig. 9). In the interim, it was decided to use the dead fish collected to examine tissue response.

When all of the dead fish from 1995 had been processed, it was determined that well over half of the test fish were already dead. We also recognized that the dead fish collected did not represent all of the dead fish. After rain storms, which would turn the tank water turbid, highly decayed fish were found that obviously could not be identified. Even with the water clear, dead fish were occasionally lost in the large tanks. With so few fish left, the experimental design was modified to continue to use mortalities to examine tissue response until mature fish could be collected in the fall 1996.

Another problem that surfaced during the winter was the presence of a freshwater copepod, *Lernaea* sp. (common name, anchor worm), on the gills of some fish. The problem become epizootic in the spring when basically any fish examined had some copepods attached to its gills. Since crustaceans are osmoconformers, it was decided to transfer the salmon to saltwater (28 ppt) net-pens in June to eliminate the copepods. Surprisingly, the copepods remained present and even sexually active in the saltwater environment. In fact, they were still present on many of the fish sampled in November 1996.

When the fish were transferred to saltwater on 14 June 1996, they were vaccinated against *Vibrio*. Each fish was also scanned for PIT tags and its fork length measured. This process revealed that unfortunately, only 208 study fish remained. The decision was made to limit our subsamples to mature fish during the falls of 1996 and 1997 in hopes of collecting some preliminary information on the tag retention of the different tag types during sexual maturation. The 1996 fall subsample was taken on 28 November to collect any mature test fish during their final stages of maturation. All fish were measured, but only mature fish were collected; the rest of the fish were left for the 1997 fall sample. Then 2 weeks later during a severe snow storm, river otters got into the net-pens and ate the remaining fish.

**Tissue response--**Since high mortalities had drastically changed the experimental design, distinctive time periods were established to examine whether tissue response changed over time. During the study, tissue response was examined over the following four periods: 1) 26 June to 31 July 1995, to cover the initial response from tagging; 2) 1 August to 31 December 1995, to cover tissue response during the first fall; 3) 1 January to 14 June 1996, to cover tissue response during the second spring; and 4) a November 1996 subsample to cover tissue response in the first mature fish.

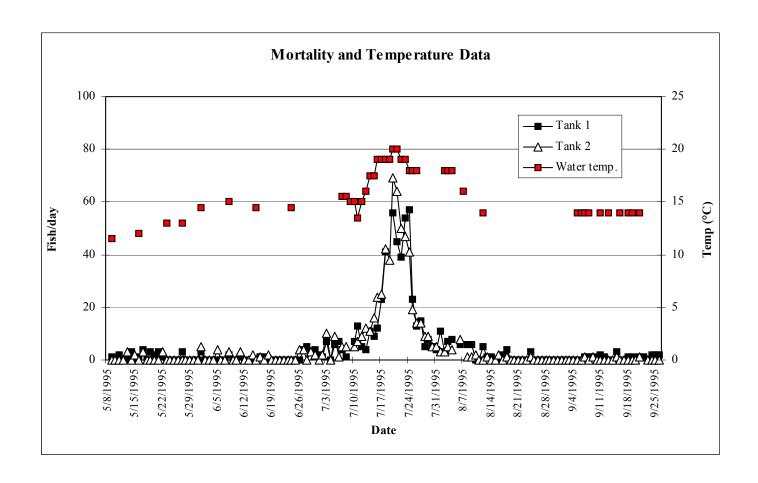


Figure 9. Number of dead coho salmon collected during spring and summer of 1995 and the Beaver Creek water temperatures recorded during the period of high mortality. The dead fish were examined for tissue response to PIT-tag treatments.

Each fish was identified as to treatment group, its fork-length measured to the nearest 1.0 mm, and its weight taken to the nearest 0.1 g. Dead tagged fish were then opened and examined for tissue response to the four tag types. Six host-response and tag-location categories were established: 1) no response, 2) encapsulated in the cecum, 3) surrounded by visceral fat, 4) attached to the intestine, 5) attached to or located inside of the air bladder, and 6) attached to the muscular wall.

If appropriate, multiple categories were used to record tissue response. Some dead fish were too decayed to determine if there had been any tissue response and therefore these were eliminated from the tissue-response analyses.

**Statistics--**Length and weight data for the five treatment groups were compared using one-way ANOVAs. After adjusting for the different numbers of fish being added to the rearing tanks for each treatment group, chi-square analyses were used to statistically evaluate mortality during the different time periods. Chi-square analyses were also used to evaluate tissue response to the four tag types. Similar numbers of fish were collected during the four sample periods for the different tag types; however, because of the possibility for individual fish to be counted under multiple categories, it was necessary to adjust the numbers so that all of the sample sizes were equal. Significance was established at  $P \le 0.05$  level.

## **Results and Discussion**

**Fish culture (growth)--**When the coho salmon were tagged, there were no significant differences in lengths (P = 0.366) or weights (P = 0.156) among the five groups (Table 13). Growth measurements were taken on all dead fish, but since the fall 1995 and spring 1996 samples were spread over months, it was not possible to use these data to compare growth among the tag types. However, length measurements were taken on all remaining fish in June, when the fish were transferred to net-pens, and during the November 1996 subsample. Similar to the tagging data, there were no significant differences in lengths among the five groups in June (P = 0.993) or November (P = 0.972; Table 14). Therefore, the tagged fish grew at the same rate as the untagged control fish. Furthermore, despite the high mortality experienced during this study, the study fish grew at typical rates for coho salmon.

**Fish culture (survival)-**-Handling and tagging, and then elevated water temperatures (> 18°C) in late July accounted for high numbers of fish being killed in the tanks (see Fig. 9). Even without tagging, handling (i.e., anesthetizing with MS-222, measuring weights and lengths) was stressful to the fish: during the first 2 weeks after tagging, the control group suffered a higher number of mortalities than the tagged groups (Table 15). However, mortality during this 2-week period was not significantly different among the five treatment groups. This mortality occurred before the high water

Table 13. Lengths and weights (averages and standard deviations) of coho salmon tagged or clipped in June 1995. The numbers of fish added to the tanks for each group are also given. P values are derived from one-way ANOVAs.

	Capped	Coated	Etched	Regular	Clipped	P
Number	486	432	493	477	508	
Length(mm)						
Average	153.4	153.4	153.6	154.6	153.2	0.366
SD	(11.7)	(9.1)	(9.3)	(15.1)	(14.0)	
Weight(g)						
Average	37.0	36.0	36.3	36.1	36.0	0.156
SD	(8.0)	(7.7)	(8.0)	(8.0)	(7.5)	

Table 14. Fork lengths (averages and standard deviations) for all coho salmon measured on 14 June and 28 November 1996. The numbers of fish remaining on these dates for each group are also given. P values are derived from one-way ANOVAs.

	Capped	Coated	Etched	Regular	Clipped	P
June1996						
Number	41	37	42	36	47	
Length(mm)						
Average	312.1	311.7	315.4	312.9	313.7	0.993
SD	(25.3)	(29.4)	(29.3)	(32.6)	(35.3)	
November1996						
Number	28	24	35	19	29	
Length(mm)						
Average	357.7	354.2	360.9	357.7	357.7	0.972
SD	(27.5)	(36.5)	(36.2)	(32.7)	(36.3)	

Table 15. Chi-square analysis of the number of mortalities recorded for the 2-week period following handling and tagging (June 26-July 10). Expected values are adjusted for the different numbers of treatment fish originally added to the tanks (see Table 13).

	Observed	Expected	$\chi^2$	
Capped	28	26.9	0.05	
Coated	27	30.2	0.34	
Etched	31	29.7	0.06	
Regular	25	31.6	1.38	
Clipped	38	30.7	1.76	
Overall	149		3.59*	

<sup>\*</sup> The significant  $\chi^2_{0.05,4}$  value is 9.49.

temperatures were recorded. Water temperature did not reach 17°C until 14 July and reached 19°C on 16 July (Fig. 9). Water temperatures remained high until early August. During this period of elevated water temperatures, large numbers of fish died in all of the treatment groups (Table 16). Again, there was no significant difference in mortality rates among groups.

An upper lethal temperature for coho salmon cannot be specifically identified because it depends on many factors; however, like other Pacific salmon, coho salmon prefer temperatures below 15°C (Bell 1991). Furthermore, elevated water temperatures have a synergistic effect and thus will be more problematic if other causes of stress are present. In this case, it appears to have been the accumulation of stresses (handling and high water temperatures) to the fish that was responsible for the high number of mortalities observed. We had originally decided to rear these fish on Beaver Creek water because other Minter Creek coho salmon had been successfully held in this water source for 2.5 years without experiencing high mortalities. To emphasize again the synergistic effect of the different stresses, neither this group of older coho salmon nor the salmon left over after tagging for this study suffered high mortality during the period of elevated water temperatures.

Using the processed dead fish, nondifferential mortality continued through 1995 (Table 17). Then using the actual number of survivors present on 28 November 1996, the data demonstrate that whatever had killed the test fish throughout the study, that all of the groups had been similarly affected (Table 18). Comparing the number of survivors and the number of dead fish collected for each group, one finds that approximately 100 fish from each group were not recovered. This represented 20-25% of the study fish.

**Tissue response through 31 July 1995.**-Using the dead fish collected through 31 July 1995, it was possible to determine that consistent tissue response occurred earlier in the Teflon-capped (11 days post-tagging) and parylene-coated (15 days post-tagging) than in the acid-etched or regular PIT-tagged (both 22 days post-tagging) groups. Furthermore, both parylene-coated and Teflon-capped groups had half as many fish as regular and etched groups that showed no tissue response (Table 19). The chi-square analysis was significant for all four groups ( $\chi^2 = 38.60$ ). Subdividing the chi-square analysis separated the four groups into two distinct groups: one combined parylene-coated and Teflon-capped fish, and the other combined acid-etched and regular PIT-tagged fish. With a standardized sample size of 191 fish per group, observed values for the acid-etched (n = 81) and regular PIT tag groups (n = 103.6) indicated that approximately half of the fish tagged in these groups had no immediate tissue response.

In examining the other categories, chi-square analysis showed statistically significant differences in every case (Table 19). In general, the Teflon-capped and parylene-coated fish continued to show more tissue response than the acid-etched and regular fish. Chi-square analysis for tags located in the ceca among the four tagged

groups was significant ( $\chi^2 = 12.07$ ), and subdivision demonstrated that the nonconformity was due primarily to the Teflon-capped fish (Table 19). Similar statistical results were found for Teflon-capped tags found surrounded by the visceral fat. After subdividing the significant chi-square analyses, the parylene-coated fish were shown primarily responsible for the nonconformity observed for tags found attached to or located inside of the air bladder, attached to the intestine, and attached to the muscular wall. Approximately 22% of the dead fish had tissue responses that were recorded under multiple categories.

Tissue response through 31 December 1995--Only the chi-square analysis for no tissue response was significant for dead fish collected during this time period (Table 20). As before, both parylene-coated and Teflon-capped groups had fewer fish than the regular and etched groups showing no tissue response. This was especially true for the Teflon-capped group, which had only 5 of 53 fish sampled showing no response. Approximately 25% of the dead fish had tissue responses that were recorded under multiple categories.

**Tissue response through 14 June 1996--**All chi-square analyses were insignificant for dead fish collected during this time period (Table 21). Although not significant, the Teflon-capped group again had the fewest number of fish with no tissue response (and therefore the highest number with some tissue response). During this time period, the sample size was standardized at 55 fish per group, with approximately 11% of the dead fish having tissue responses that were recorded under multiple categories.

**Tissue response in November 1996 subsample-**-This time period had the smallest standardized sample size (14 fish/group); however, these were the mature fish that the study was design to examine. Similar to the two earliest time periods, the chi-square analysis was significant for the no tissue response category (Table 22). Also like before, this was primarily because of the high number of regular PIT-tagged fish in this category. Furthermore, the Teflon-capped group again had the fewest number of fish with no tissue response.

There were two mature fish that were not clipped and did not have PIT tags; these probably had lost their tags. One was a male and one was a female. Unfortunately, it was impossible to tell which group they came from. If one were repeating this study, we recommend that study fish be double tagged with a batch tag so that one could at least identify the treatment group on fish that lost their PIT tags. One could use coded-wire tags or photonic tags for this purpose. This would be a better solution than rearing the fish in separate containers.

Although the study ended prematurely, preliminary results do indicate that fish tagged with Teflon-capped PIT tags appear to have more tissue response than those tagged with regular PIT tags. However, we still do not know if this tissue response will

Table 16. Chi-square analysis of the number of mortalities recorded between 15 July and 31 July 1995 when water temperatures were high. The expected values are adjusted for the different numbers of treatment fish originally added to the tanks (see Table 13).

	Observed	Expected	$\chi^2$	
Capped	168	179.9	0.79	
Coated	146	159.9	1.21	
Etched	193	176.6	1.53	
Regular	197	188.1	0.42	
Clipped	183	182.5	0.00	
Overall	887		$3.95^{*}$	

<sup>\*</sup> The significant  $\chi^2 2_{0.05,4}$  value is 9.49.

Table 17. Chi-square analysis of the number of mortalities recorded between 1 August and 31 December 1995. The expected values are adjusted for the different numbers of treatment fish originally added to the tanks (see Table 13).

	Observed	Expected	$\chi^2$	
Capped	66	57.0	1.42	
Coated	61	50.7	2.11	
Etched	43	55.9	2.99	
Regular	45	59.6	3.57	
Clipped	66	57.8	1.16	
Overall	281		$7.68^{*}$	

<sup>\*</sup> The significant  $\chi^2_{0.05,4}$  value is 9.49.

Table 18. Chi-square analysis of the number of mortalities for the entire study based on actual numbers of fish left on 28 November 1996. The expected values are adjusted for the different numbers of treatment fish added to the tanks (see Table 13).

	Observed	Expected	$\chi^2$	
Capped	458	458.6	0.00	_
Coated	408	407.7	0.00	
Etched	458	450.1	0.14	
Regular	458	479.4	0.95	
Clipped	479	465.2	0.41	
Overall	2261		1.50*	

<sup>\*</sup> The significant  $\chi^2_{0.05,4}$  value is 9.49.

Table 19. Chi-square analysis of the tissue responses for the four tag treatment groups through 31 July 1995. The observed values are adjusted as if all four groups had 191 fish sampled. Asterisks designate significant deviations from the expected number of individuals to have that response.<sup>a</sup>

	Capped	Coated	Etched	Regular	Totals
No response					
Observed	40.6	47.1	81.0	103.6	272.3
Expected	68.1	68.1	68.1	68.1	
$\chi^2$	11.3	6.4	2.4	18.6	$38.60^*$
Cecum					
Observed	18.1	11.2	6.0	4.1	39.4
Expected	9.8	9.8	9.8	9.8	
$\chi^2$	7.0	0.2	1.5	3.4	$12.07^{*}$
Visceral fat					
Observed	35.2	22.3	25.0	15.2	97.8
Expected	24.4	24.4	24.4	24.4	
$\chi^2$	4.7	0.2	0.0	3.5	8.41*
Intestine					
Observed	59.8	71.9	48.0	40.6	220.3
Expected	55.1	55.1	55.1	55.1	
$\chi^2$	0.4	5.2	0.9	3.8	10.25*
Air bladder					
Observed	55.5	78.1	49.0	40.6	223.3
Expected	55.8	55.8	55.8	55.8	
$\chi^2$	0.0	8.9	0.8	4.1	13.89*
Muscular wall					
Observed	27.7	39.7	20.0	18.3	105.7
Expected	26.4	26.4	26.4	26.4	
$\chi^2$	0.7	6.6	1.6	2.5	10.79*

 $<sup>^{</sup>a}$  The significant  $\chi^{2}_{0.05,3}$  value is 7.815.

Table 20. Chi-square analysis of the tissue responses for the four tag treatment groups from 1 August through 31 December 1995. The observed values are adjusted as if all four groups had 53 fish sampled. Asterisks designate significant deviations from the expected number of individuals to have that response.<sup>a</sup>

	Capped	Coated	Etched	Regular	Totals
No response					_
Observed	5.0	14.1	25.7	32.1	76.8
Expected	19.2	19.2	19.2	19.2	
$\chi^2$	0.5	1.4	2.2	8.7	22.73*
Cecum					
Observed	6.0	2.2	1.7	0.0	9.9
Expected	2.5	2.5	2.5	2.5	
$\chi^2$	5.0	0.0	0.2	2.5	7.79
Visceral fat					
Observed	20.0	11.9	13.7	12.8	58.4
Expected	14.6	14.6	14.6	14.6	
$\chi^2$	2.0	0.5	0.1	0.2	2.76
Intestine					
Observed	16.0	16.2	10.3	6.4	48.9
Expected	12.2	12.2	12.2	12.2	
$\chi^2$	1.2	1.2	0.3	2.8	5.54
Air bladder					
Observed	18.0	17.3	6.8	9.6	51.8
Expected	12.9	12.9	12.9	12.9	
$\chi^2$	2.0	1.5	2.9	0.8	7.17
Muscular wall					
Observed	4.0	7.6	6.8	3.2	21.6
Expected	5.4	5.4	5.4	5.4	
$\chi^2$	0.4	0.9	0.4	0.9	2.50

 $<sup>^{\</sup>text{a}}$  The significant  $\chi^2_{0.05,3}$  value is 7.815.

Table 21. Chi-square analysis of the tissue responses for the four tag treatment groups from 1 January through 14 June 1996. The observed values are adjusted as if all four groups had 55 fish sampled. Asterisks designate significant deviations from the expected number of individuals to have that response.<sup>a</sup>

	Capped	Coated	Etched	Regular	Totals
No response					
Observed	24.0	33.0	32.2	29.8	119.0
Expected	29.8	29.8	29.8	29.8	
$\chi^2$	1.1	0.4	0.2	0.0	1.67*
Cecum					
Observed	4.0	2.7	0.0	3.4	10.2
Expected	2.5	2.5	2.5	2.5	
$\chi^2$	0.8	0.0	2.6	0.3	3.70
Visceral fat					
Observed	9.0	6.9	11.4	6.9	34.1
Expected	8.5	8.5	8.5	8.5	
$\chi^2$	0.0	0.3	0.9	0.3	1.62
Intestine					
Observed	12.0	12.4	9.5	5.7	39.6
Expected	9.9	9.9	9.9	9.9	
$\chi^2$	0.4	0.6	0.0	1.8	2.84
Air bladder					
Observed	7.0	4.1	3.8	10.3	25.2
Expected	6.3	6.3	6.3	6.3	
$\chi^2$	0.1	0.8	1.0	2.5	4.38
Muscular wall					
Observed	5.0	4.1	3.8	2.3	15.2
Expected	3.8	3.8	3.8	3.8	
$\chi^2$	0.4	0.0	0.0	0.6	1.00

 $<sup>^{</sup>a}$  The significant  $\chi^{2}_{0.05,3}$  value is 7.815.

Table 22. Chi-square analysis of the tissue responses for the four tag treatment groups for the November 1996 subsample of mature fish. The observed values are adjusted as if all four groups had 14 fish sampled. Asterisks designate significant deviations from the expected number of individuals to have that response.<sup>a</sup>

	Capped	Coated	Etched	Regular	Totals
No response					
Observed	1.1	1.3	3.0	9.3	14.7
Expected	3.7	3.7	3.7	3.7	
$\chi^2$	1.8	1.6	0.1	8.7	12.26*
Cecum					
Observed	0.0	0.0	0.0	2.3	2.3
Expected	0.6	0.6	0.6	0.6	
$\chi^2$	0.6	0.6	0.6	5.2	7.00
Visceral fat					
Observed	0.0	1.3	0.0	2.3	3.6
Expected	0.9	0.9	0.9	0.9	
$\chi^2$	0.9	0.2	0.9	2.3	4.23
Intestine					
Observed	5.4	3.8	6.0	0.0	15.2
Expected	3.8	3.8	3.8	3.8	
$\chi^2$	0.7	0.0	1.3	3.8	5.73
Air bladder					
Observed	3.2	1.3	0.0	0.0	4.5
Expected	1.1	1.1	1.1	1.1	
$\chi^2$	3.9	0.0	1.1	1.1	6.21
Muscular wall					
Observed	3.2	0.0	3.0	0.0	6.2
Expected	1.6	1.6	1.6	1.6	
$\chi^2$	1.8	1.6	1.3	1.6	6.25

<sup>&</sup>lt;sup>a</sup> The significant  $\chi^2_{0.05,3}$  value is 7.815.

translate into better tag retention during sexual maturation. In addition, preliminary results indicate it would be possible to remove etched PIT tags from the study because their results so closely resembled results for the regular PIT tags.

#### **Conclusions and Recommendations**

Growth and survival results were not significantly different among the five treatment groups at any time during the entire study. However, using the dead fish collected through 31 July 1995, it was possible to determine that consistent tissue response occurred earlier in the Teflon-capped (11 days post-tagging) and parylene-coated (15 days post-tagging) than in the acid-etched or regular PIT-tagged (both 22 days post-tagging) groups. Furthermore, both Teflon-capped and parylene-coated fish had half as many fish as etched and regular groups showing no tissue response during this first time period.

In all four of the time periods, the most consistent trend was that the regular PIT-tag group had the highest number of fish with no tissue response and the Teflon-capped group had the highest number with some tissue response. However, we still do not know if this tissue response will translate into better tag retention by the Teflon-capped fish during sexual maturation.

With the CRB recovery plans requiring the development of PIT-tag interrogation systems for adult salmon, NMFS recommends that this experiment be repeated. However, there are a few fish culture changes that NMFS recommends if this experiment were to be repeated. We recommend that the tagging be done in early spring before water temperatures begin to rise. We also recommend that weights be taken on only 10% of the study fish instead of the 100%, because it is necessary to anesthetize fish longer when weights are being taken than if one is only tagging and taking lengths. Finally, we recommend that smaller tanks be used so that it is easier to find the dead fish and that study fish be double tagged with a batch tag so that one could at least identify the treatment group on fish that have lost their PIT tags.

## ACTIVITIES AT COLUMBIA RIVER BASIN DAMS

# **Review of PIT-tag Systems**

Fisheries agencies have requested that PIT-tag interrogation systems and general sampling facilities for juvenile salmon be constructed at Ice Harbor, John Day, The Dalles, and Bonneville Dams over the next 5 years (see Fig. 1). In addition, they requested that the juvenile fish bypass/collection facility at Lower Granite Dam be upgraded. The agencies also requested that the new facilities include PIT-tag separation systems. During 1994-1995, NMFS worked with the COE and its contractors in reviewing engineering concept drawings for these facilities. This review led to changes in number and placement of several interrogation units, electrical specifications, and water-flow requirements. NMFS will continue to consult with the COE and its contractors regarding technical matters related to the location and installation of PIT-tag and related systems at the new facilities.

In August 1994, NMFS personnel joined a team of biologists from several fisheries agencies and the COE in reviewing future PIT-tag interrogation and fish separation needs for Lower Granite, Little Goose, Lower Monumental, and McNary Dams (Fig. 1). The team's recommendations were presented to BPA in late 1994. BPA approved the installation of interrogation units for monitoring mortalities at these dams; however, BPA did not support the relocation of existing interrogation units or installation of new interrogation units on the river and barge exits.

# **Installation of PIT-tag Systems**

During 1994, new bypass/collection facilities for juvenile salmon were completed at McNary and Lower Monumental Dams. The basic McNary facility, which included PIT-tag interrogation and slide-gate separation systems, was built by the COE. PSMFC installed the electronic components and cabling for the interrogation and separation systems. Personnel from NMFS acted as advisors to PSMFC staff and assisted them with installation. The McNary facility became operational on schedule, in April 1994.

Construction of the new bypass/collection facility at Lower Monumental Dam, which included its PIT-tag interrogation and separation systems, was scheduled to be completed in early 1993 by the COE. When the facility was not completed on time, NMFS installed a temporary PIT-tag interrogation system in spring 1993. Permanent PIT-tag interrogation and separation systems were installed by NMFS staff with assistance from PSMFC before the 1994 field season.

In 1995, an experimental site at Lower Granite Dam (GRX) was established as a platform for evaluating the new rotational fish diversion gates and the computer program (BYCODE) that controls fish separation. NMFS installed prototype two-way and three-way rotational gates, six dual-coil interrogation units, and all of the necessary electronic and computer equipment. The GRX site operated independently of the main site at Lower Granite Dam (GRJ) and was designed to divert PIT-tagged fish to the river or to a series of three holding tanks.

The rotational gates and BYCODE program were successfully evaluated in 1995 and the GRX site has been used by many researchers since its construction. A similar experimental site (GOX) was established at Little Goose Dam in 1996. At GOX, NMFS installed a two-way rotational gate and a two-way side-to-side gate. They also installed a secondary fish holding tank into the large tank that was already on site. Like GRX, this site operates independent of the main GOJ site and is used by researchers to collect their study fish.

# **Measurements of Radio-Frequency Emissions**

## Introduction

Radio frequency emissions from PIT-tag equipment must comply with Federal Communications Commission (FCC) and National Telecommunications and Information Administration (NTIA) regulations for low-power electronics equipment. The FCC and NTIA regulations indicate that with 400-kHz equipment, RF emissions must be below 6 FV/m when measured at 300 m from several locations. Extrapolation, using the inverse distance squared, is permitted if 300 m cannot be directly measured. In 1994, RF emissions were measured at Little Goose, McNary, and Lower Monumental Dams.

## **Methods and Materials**

Each dual-coil interrogation unit was measured independently. The first step was to find locations where measurements could be made that were approximately 300 m from the PIT-tag interrogation unit. The exact distance of the location was then determined using either triangulation or a laser range finder. Emissions were measured using a calibrated spectral analyzer (Hewlett-Packard model 3585A) set at 400 kHz and its harmonic frequencies. The spectral analyzer was connected to a calibrated loop antenna (Antenna Research Associates model BBH-1100/A) that was rotated to determine maximum emission strength.

## **Results and Discussion**

At Little Goose Dam, four dual-coil interrogation units had exceeded the 6 FV/m limit for RF emissions in 1993. Between the 1993 and 1994 seasons, new aluminum shields were fabricated for these units. Originally, in March 1994, two of the interrogation units still did not comply. Upon closer inspection, it was determined the problem was in the dual-exciter boards. Loops on the exciter boards determine the direction the magnetic fields flow through the coils. Normally, the loops are set to make the fields flow in opposite directions, which minimizes the RF emissions. Instead the two failing units had their coils' fields flowing in the same direction, which enhanced the fields and resulted in higher RF emissions. Once the loops were reversed, the two units complied with the regulations.

When measurements were done at McNary Dam, all except one dual-coil interrogation unit exceeded the 6 FV/m limit for RF emissions. When the shields were inspected, it was obvious their seams were not electrically connected (i.e., not welded together). The noncontinuous seams permitted the RF emissions to escape easily from the shields. These shields had been fabricated by a COE contractor and had not been built according to NMFS design specifications. In contrast, all of the measurements were below the 6 FV/m limit at Lower Monumental Dam where the shields were fabricated using NMFS specifications. Based on this finding, all PIT-tag interrogation system shields at McNary Dam were modified by the COE. Therefore, NMFS recommends that all future installation of PIT-tag interrogation systems require shields that meet the NMFS design specifications.

# **Performance of Fixed-Reference Tags**

In 1993, personnel from the NMFS Sand Point Electronics Shop developed a fixed-reference tag to use as a maintenance tool for PIT-tag interrogation systems (Prentice et al. 1994). The fixed-reference tags test the operational status of each excitation/detection coil by simulating the passage of two PIT tags through that particular coil. They are set to activate their two tags every 4 hours, and the transmitted tag codes become part of the permanent PTAGIS computer file. If a tag code is not recorded, this indicates a potential problem in the interrogation system.

This is especially useful when few fish are passing through the bypass/collection facilities. For example, without the fixed-reference tag information, if a PIT-tag code had not been recorded for hours or days, then one would not know whether the coil was defective or no PIT-tagged fish had transited the flume. Thus, the ability to determine the operational status of each coil on a daily basis is important from systems-reliability and data-integrity standpoints.

During February and March 1994, fixed-reference tags were installed on each coil of the interrogation systems at Lower Granite, Little Goose, Lower Monumental, McNary, and Prosser Dams. To maximize the usefulness of the fixed-reference tags, PSMFC personnel developed a computer program to separate fixed-reference tag data from normal PIT-tag data. The computer program generates an observation-summary report for the fixed-reference tag codes based on the previous day's data. In this report, if a tag code was not received during any of its six transmissions, the potentially defective coil was listed for immediate attention. Maintenance personnel from PSMFC then investigated the situation and corrected any problems.

During the 1994 field season, PIT-tag interrogation systems at the dams experienced only a few electronically related problems. In several cases, the fixed-reference tags were critical in alerting maintenance personnel of coil failures that would otherwise have gone undetected. For example, there was a problem at Lower Granite Dam with a coil located in a flume that was inactive at that time. Even in the few cases when active coils failed, because of the fixed-reference tags, they were repaired quickly. These examples demonstrate that this new tool has significantly improved the overall maintenance and trouble-response time.

In 1995, NMFS requested that Destron-Fearing modify the tag codes of the fixed-reference tags so that all started with the common four-letter code, 0B0B. The change enabled fixed-reference tag codes to be easily identified from normal PIT-tag codes in the computer file. This change helped to improve on-site system analysis, because problems were detected immediately without having to wait for the observation-summary report that would be listing yesterday's problems. The fixed-reference tags operated as designed during the 1996 field season. Furthermore, fixed-reference tags have become a critical maintenance tool for PSMFC.

# **Evaluation of the Separation-by-Code System at Lower Granite Dam**

## Introduction

During 1992-1994, NMFS developed and evaluated the Separation-by-Code system at the Manchester Research Station. A Separation-by-Code system combines the computer program, BYCODE, with one or more fish diversion gates. The Separation-by-Code system uses the computer program to separate targeted PIT-tagged fish from untargeted tagged and untagged fish based on their individual tag codes. BYCODE sends a signal to a fish diversion gate when it wants a particular fish diverted. By fall 1994, the Separation-by-Code system had successfully passed its tests at Manchester. To start the transfer of this technology from the research and development stage at NMFS to the operations and maintenance environment at PSMFC, it was necessary to evaluate the system at a dam. Therefore, the experimental site at Lower Granite Dam, GRX, was established.

During the spring of 1995, NMFS installed two prototype rotational gates, interrogation units, computers, and all of the related electronic hardware. Then, BYCODE was evaluated for its ability to direct PIT-tagged fish into five pathways, and the rotational gates were evaluated for mechanical performance. To determine how fish behavior and fish density affected gate efficiencies, tests were conducted in April (low fish density) and May (high fish density) using two salmon species.

## **Methods and Materials**

At the first fish diversion gate (two-way rotational) at GRX, fish can either continue to the river or be diverted toward the second diversion gate (Fig. 10). At the second diversion gate (three-way rotational), fish can continue down the center pathway or be diverted left or right. Net-pens for collecting fish were installed at the ends of these three pathways. The collected fish enabled separation efficiencies to be calculated. For each test, separation efficiencies were calculated for these five possible pathways: to the river and to the three-way diversion gate for the two-way gate; and to the center, left, and right directions for the three-way gate.

Spring chinook salmon (*Oncorhynchus tshawytscha*; n = 500) and steelhead (*O. mykiss*; n = 500) were tested in April when only a few juvenile fish were migrating through the dam. Fall chinook salmon (n = 500) and steelhead (n = 500) were tested in May when large numbers of salmon were migrating through Lower Granite Dam. In the computer database for each species, fish were divided equally among the four final destinations (i.e., 125 fish to the river, center net, left net, and right net). This was done by assigning Action codes to individual tag codes within the Tag Database file used by BYCODE.

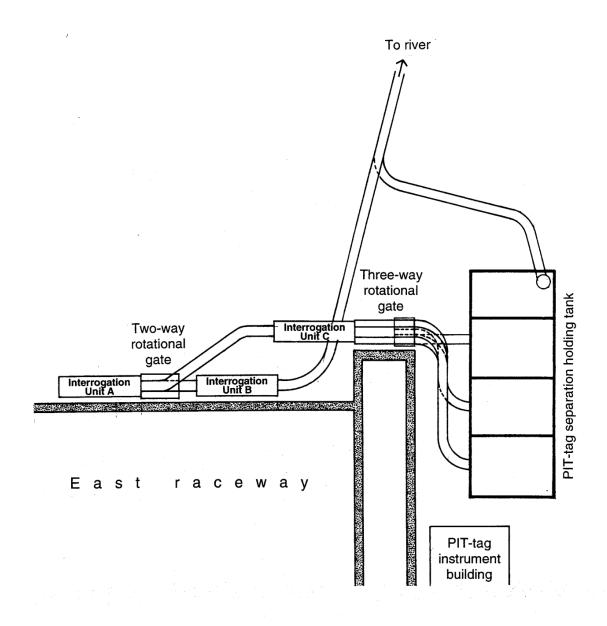


Figure 10. Diagram of the experimental site at Lower Granite Dam (GRX).

Test fish were added to the PIT-tag head tank so they went through GRX with the regular PIT-tagged fish. Non-study PIT-tagged fish had their tag codes recorded by the computer program, but because they were not programmed to be diverted, they should have all gone to the river. However, if they were traveling closely in front of or behind a test fish, they could have been diverted with that test fish.

## **Results and Discussion**

Separation efficiencies for the five pathways for spring and fall chinook salmon ranged from 93.5% to 96.8% and 90.2% to 100%, respectively (Table 23). Although they had similar separation efficiencies, the two run types behaved differently: fall chinook salmon only migrated through GRX between dusk and dawn, while the spring chinook migrated almost immediately after they were added to the PIT-tag head tank.

Unlike chinook salmon, separation efficiencies for steelhead were notably lower for particular pathways. At the two-way gate in April, 94.1% of the steelhead assigned to the three nets were successfully diverted; however, only 73.3% of the steelhead assigned to continue to the river made it to the river. At the three-way gate, 92.3% of the fish assigned to the center net were recovered there, while only 76.8% and 79.9% were successfully diverted left and right, respectively. Steelhead appeared to react (by swimming in the flume) to the hydraulic changes present at the rotational gates.

To counteract the swimming behavior, BYCODE was modified to permit setting different delay and open times for each species at each gate. Opening the gate longer (1200 milliseconds compared to 1000 milliseconds) for steelhead increased separation efficiency for the river-assigned fish from 73.3 to 89.7% (Fig. 11). Unfortunately, because the water velocity was only around 1-1.5 m/second at the three-way gate compared to almost 3 m/second at the two-way gate, there was not a similar increase for the left- and right-assigned fish (i.e., efficiencies remained below 80%). In addition, the flume section immediately preceding the three-way gate has a sharp Z turn in it, which appeared to start the fish responding even before they reached the gate.

In April, only three non-study PIT-tagged fish were recorded, and all went successfully to the river. In May, of the approximate 600 non-study PIT-tagged fish recorded, 30 fish were diverted along with study fish to the three-way gate. No untagged fish were recovered in the nets in April and only nine were recovered in May.

In general, the prototype rotational gates performed satisfactorily. However, it was observed during May that the rotational speed of the gates had slowed down relative to speeds observed in the April tests. Their delay and open settings were adjusted to accommodate the slower gates. The gates had probably slowed down from debris collecting in their mechanisms.

Table 23. Separation efficiencies (%) for the five pathways for spring and fall chinook salmon.

	Spring chinook salmon	Fall chinook salmon
Two-way gate		
To river	93.5	90.5
To three-way gate	96.7	99.3
Three-way gate		
Left	96.6	93.4
Center	96.8	100.0
Right	96.2	90.2

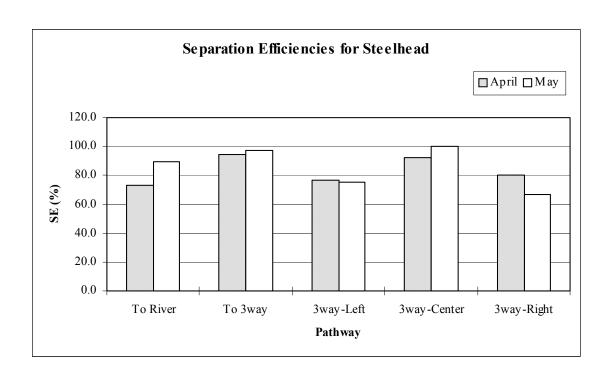


Figure 11. Separation efficiencies for the five different pathways for both the April and May tests for steelhead at Lower Granite Dam. The open times for the gates were 1000 milliseconds in April and 1200 milliseconds in May.

## **Conclusions and Recommendations**

Evaluation of the Separation-by-Code system at GRX was highly successful. For the most part, 90-100% of the targeted fish were successfully routed to their final destinations. The low water velocity at the three-way gate allowed steelhead to avoid being diverted by the gate. Therefore, NMFS recommends that any designs for future bypass systems ensure that water in all flumes associated with fish separation flows at 3-4 m/second.

In general, the prototype rotational gates performed satisfactorily. However, it was observed during May that the rotational speed of the gates had slowed down relative to speeds observed in the April tests. Their delay and open settings were adjusted to accommodate the slower gates. The gates had probably slowed down from debris collecting in their mechanisms.

Due to a short delivery schedule, there were several areas where deficiencies in BYCODE were permitted in order to meet the basic goal of installation and testing at Lower Granite Dam in spring 1995. We recommend that these deficiencies (e.g., a procedure for automatically switching to the backup computer when the primary computer fails) be completed before the system is transferred to PSMFC. An updated version of the computer program was used by four research projects during 1996 at Lower Granite Dam. These projects, along with the research projects at GOX, suggested a few more modifications that would improve the program. These changes will be completed before the 1997 season and thus will be included in the version PSMFC plans to use at most of its sites in 1997.

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